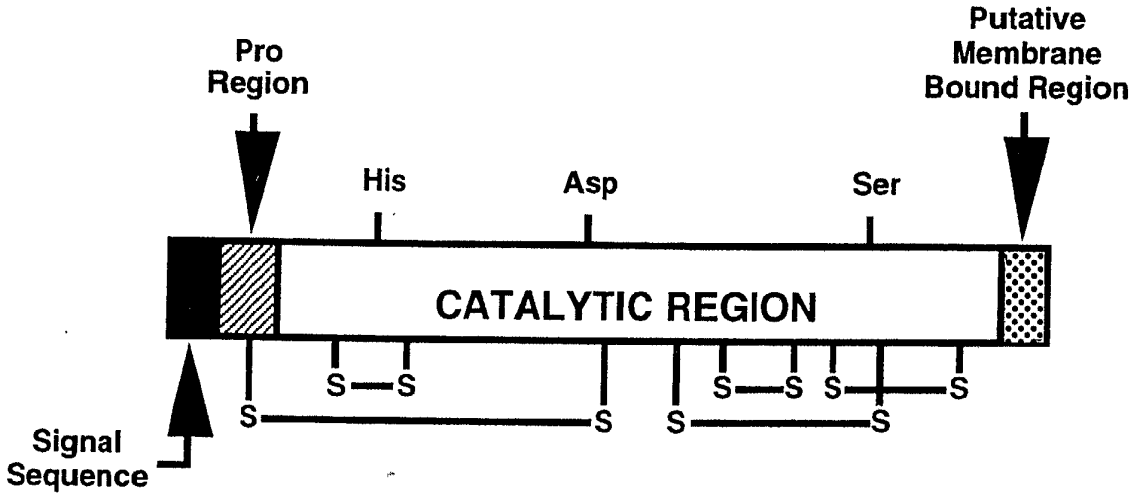
FIG 1


Pro Region
Putative Membrane Bound Region
His
Asp
Ser
CATALYTIC REGION
S—S
S—S
S—S
S—S
S—S
S—S
Signal Sequence

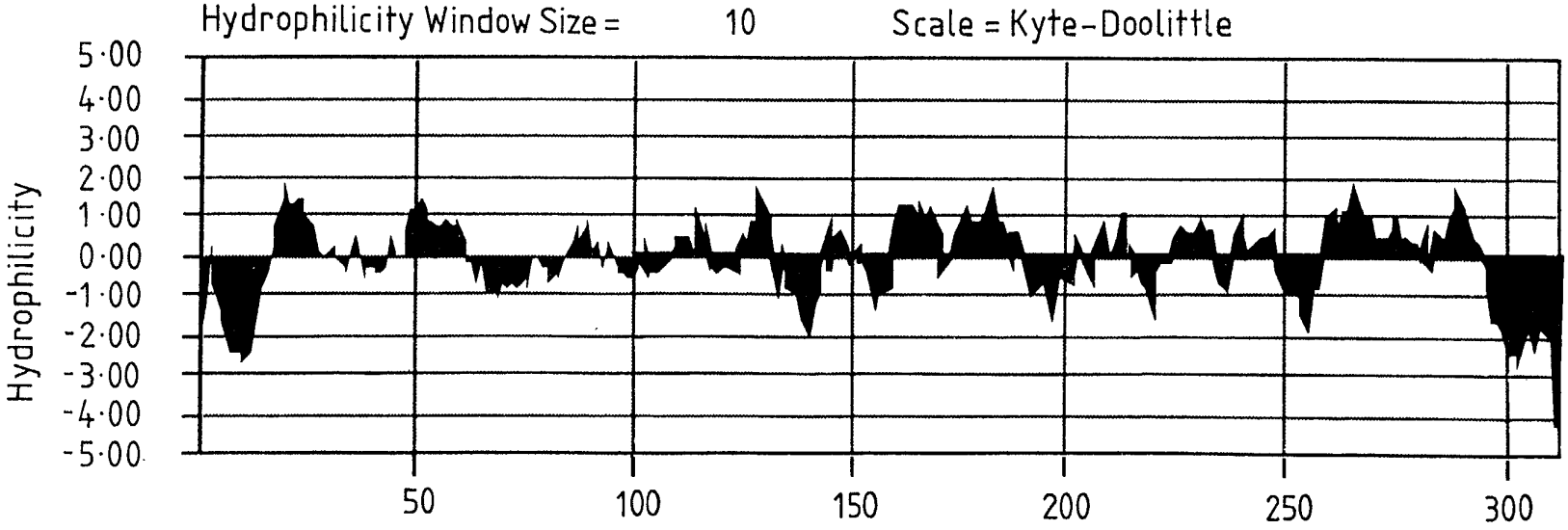
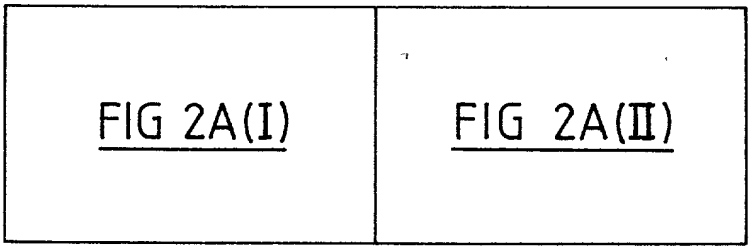

Hydrophilicity Window Size = 10
Scale = Kyte-Doolittle
Hydrophilicity
5·00
4·00
3·00
2·00
1·00
0·00
-1·00
-2·00
-3·00
-4·00
-5·00
50
100
150
200
250
300

FIG 2A



Sequence comparison of HELA2(Testisin) and prostasin

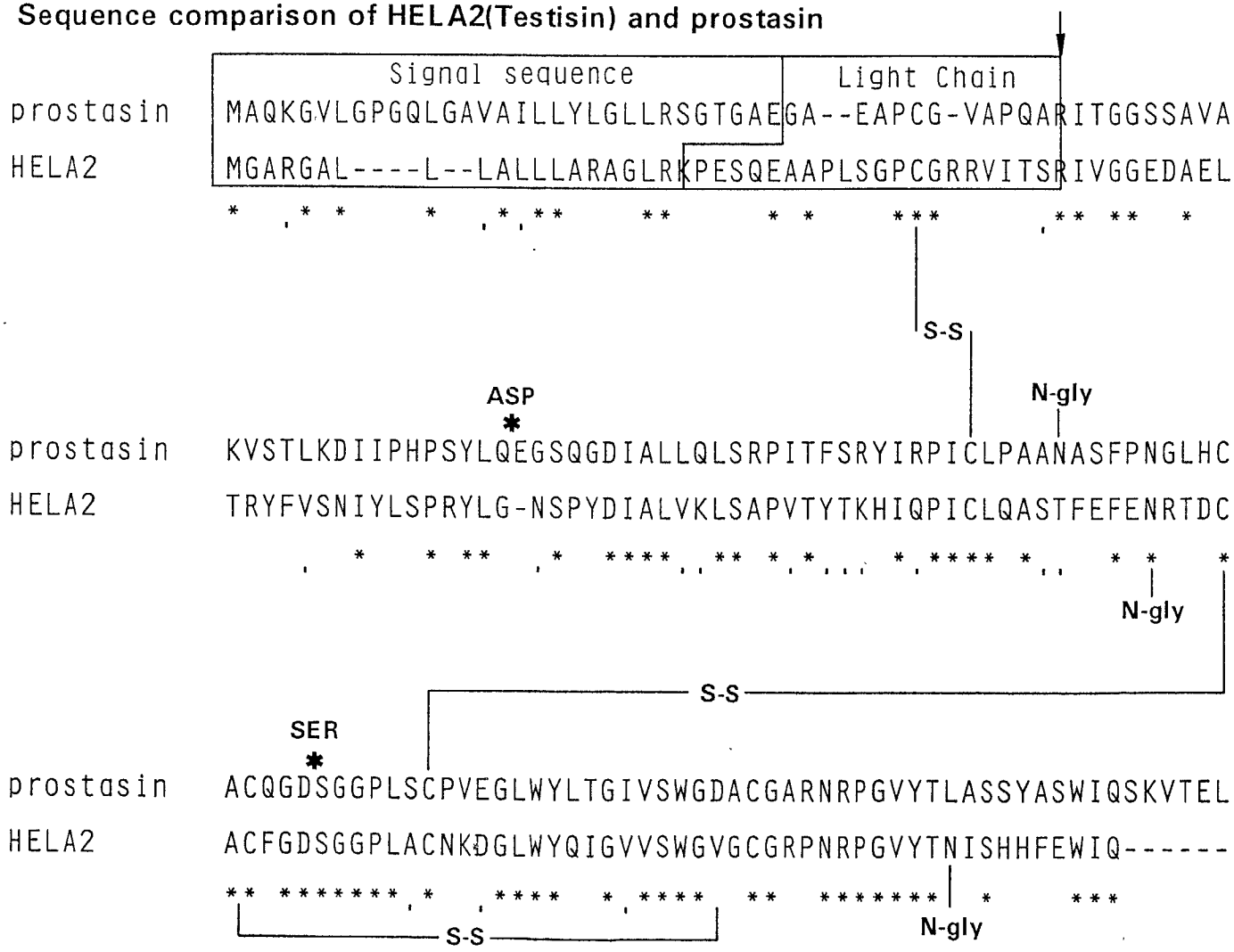


FIG 2A(I)

Sequence comparison of HELA2(Testisin) and prostasin

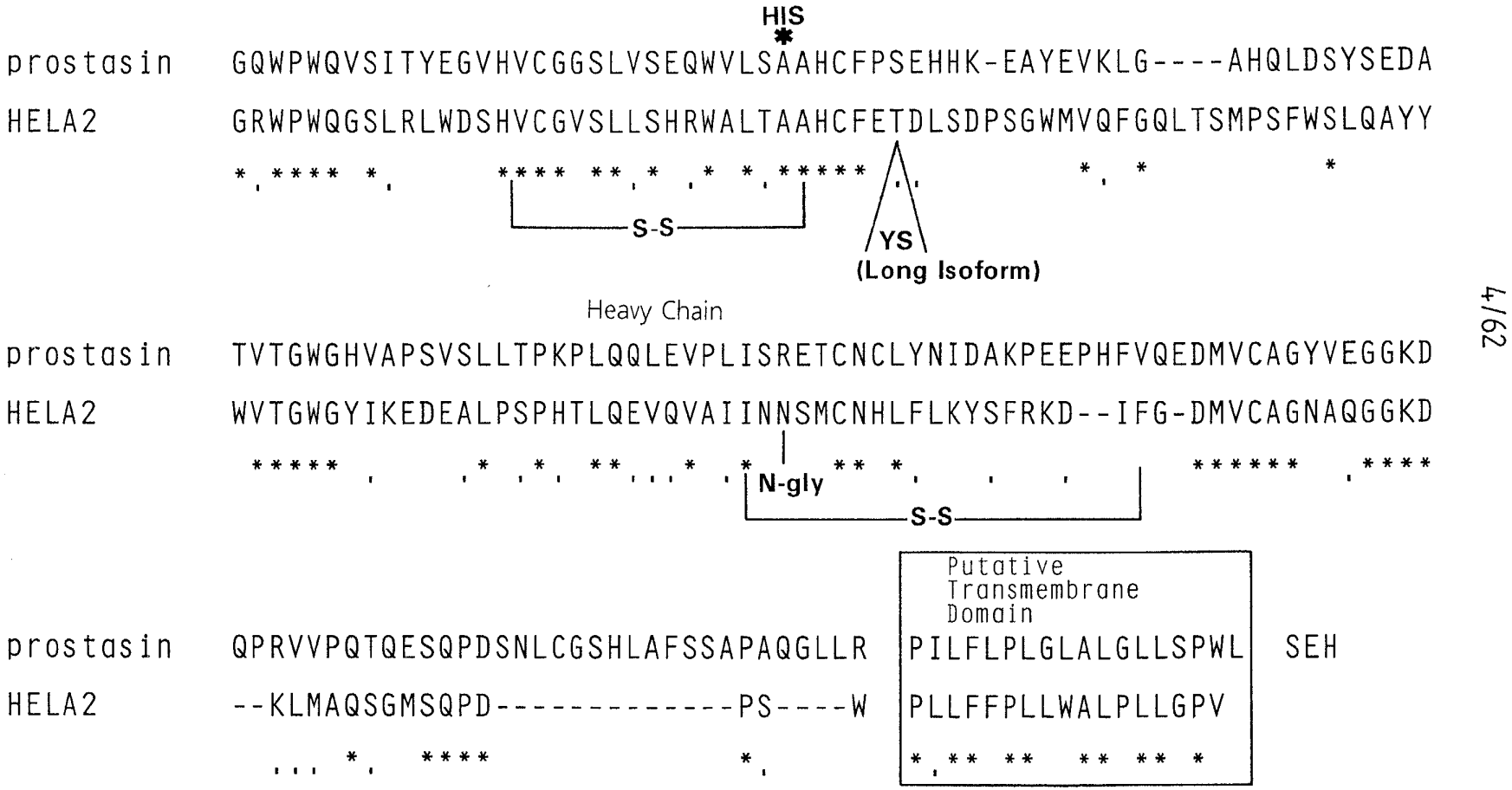
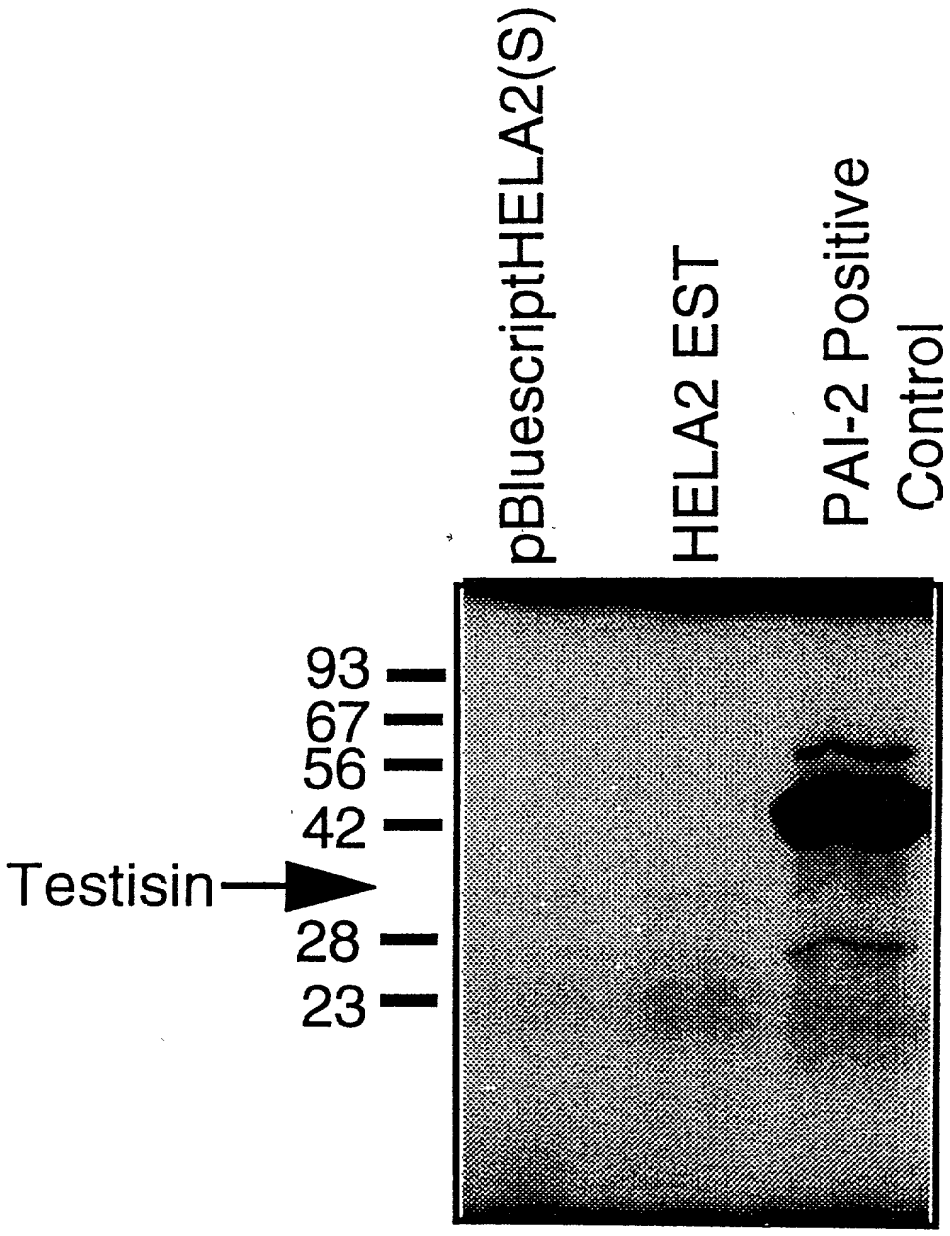


FIG 2A(II)

FIG 2B



In vitro transcription / translation of HELA2 (Testisin)

FIG 3

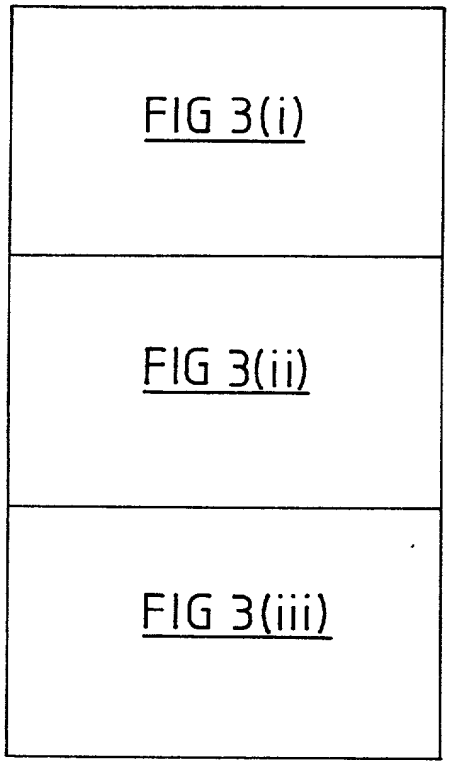
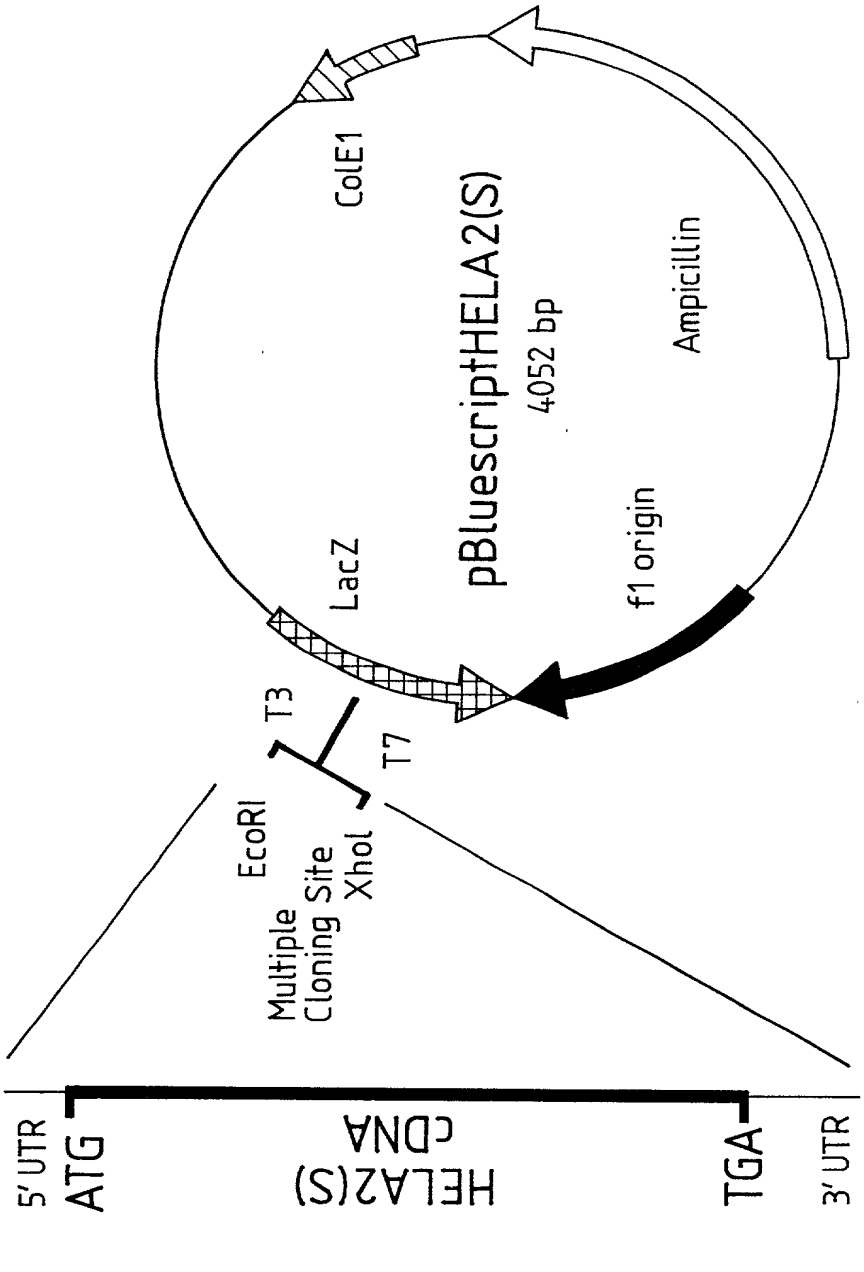
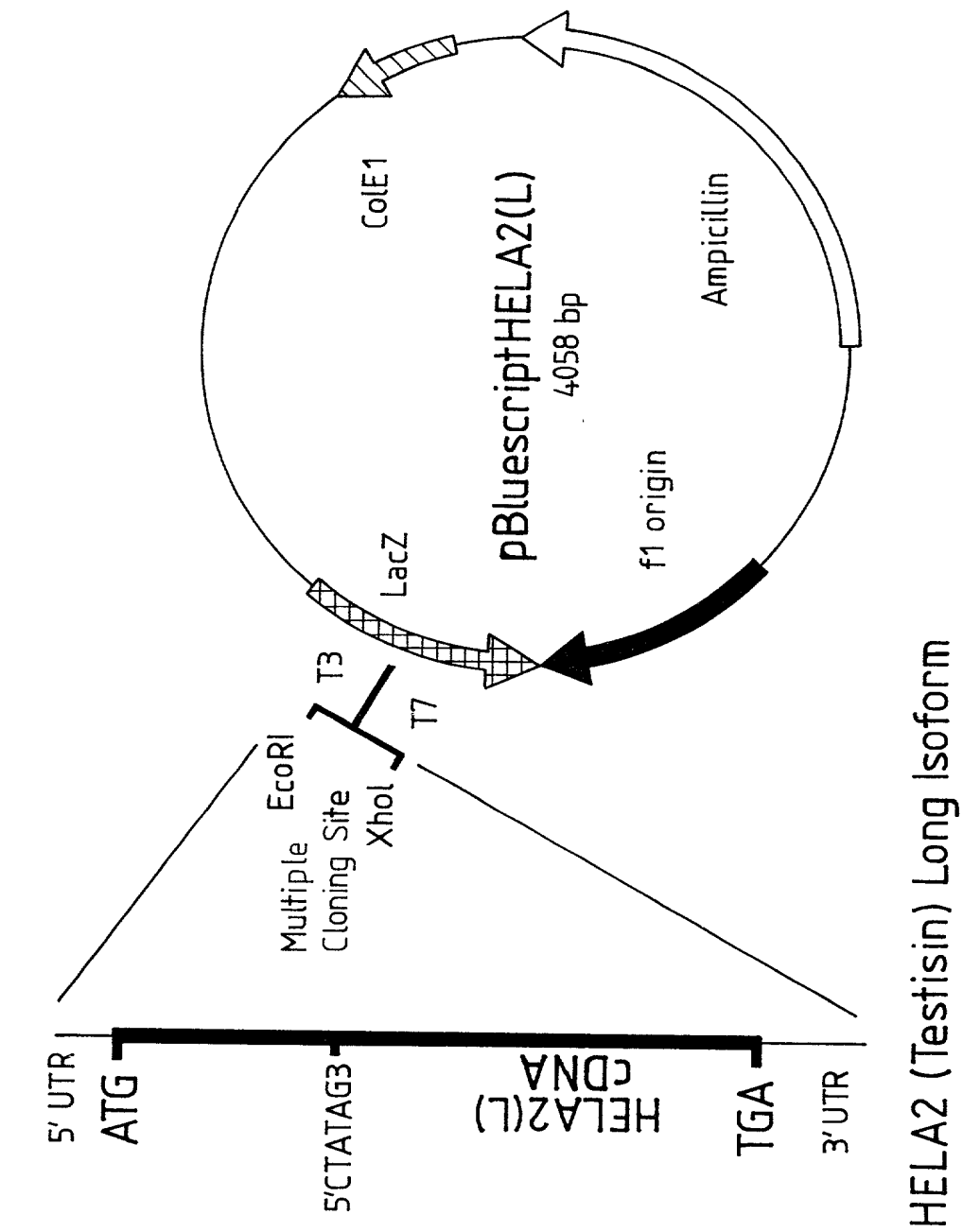


FIG 3(i)



HELA2 (Testisin) Short Isoform

FIG 3(ii)


pBluescriptHELA2(L)
4058 bp
ColE1
Ampicillin
f1 origin
LacZ
T3
T7
EcoRI
XhoI
Multiple Cloning Site
5'CTATAG3
5' UTR
ATG
HELA2(L) cDNA
TGA
3'UTR
HELA2 (Testisin) Long Isoform

HELA2 (Testisin) Restriction Enzyme Map

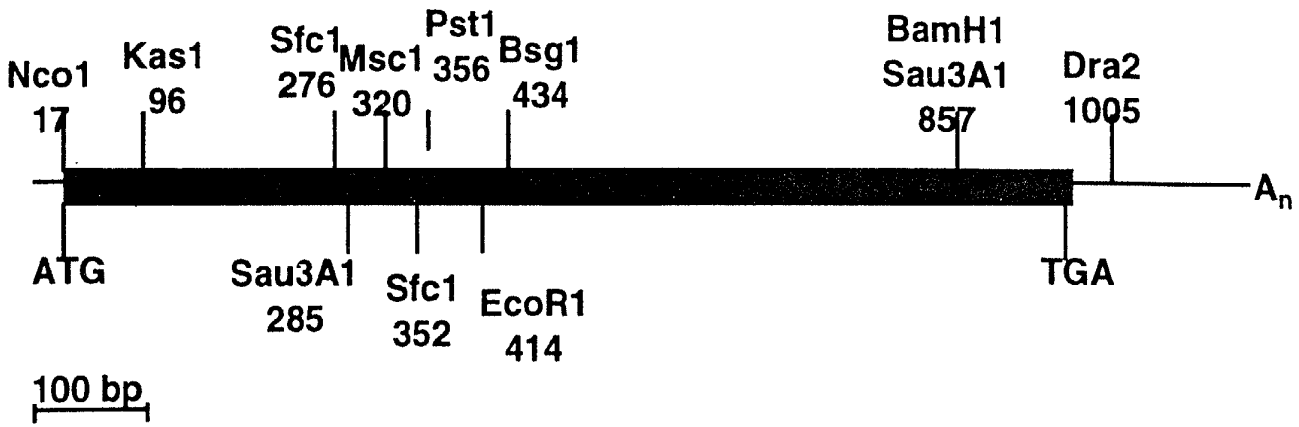
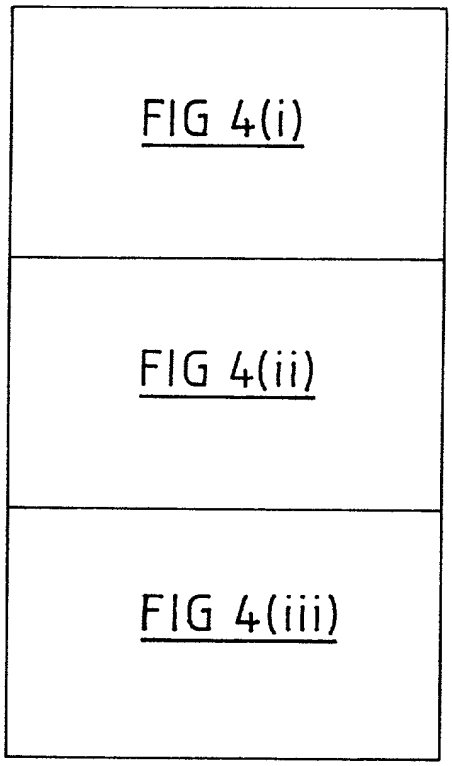


FIG 3(iii)

FIG 4



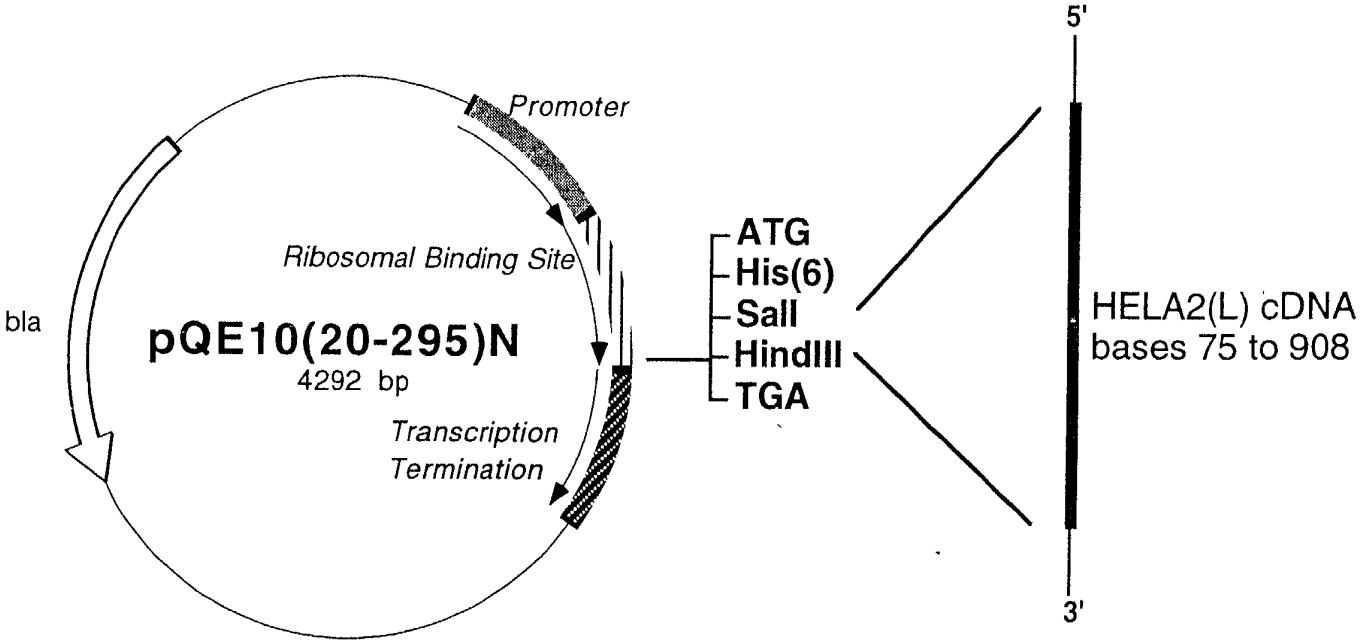

5'
Promoter
Ribosomal Binding Site
bla
pQE10(20-295)N
4292 bp
Transcription
Termination
ATG
His(6)
SalI
HindIII
TGA
HELA2(L) cDNA
bases 75 to 908
3'

FIG 4(i)

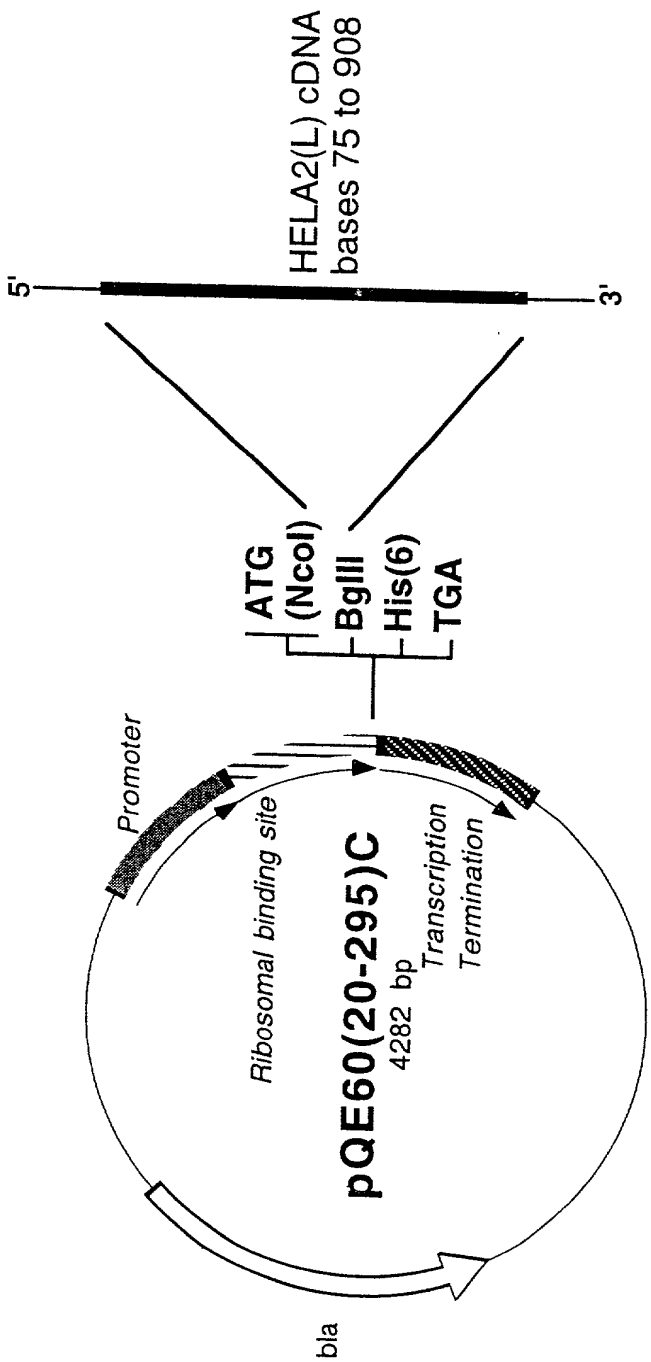

HELA2(L) cDNA
bases 75 to 908
5'
3'
ATG
(NcoI)
BglII
His(6)
TGA
Promoter
Ribosomal binding site
pQE60(20-295)C
4282 bp
Transcription
Termination
bla

FIG 4(ii)

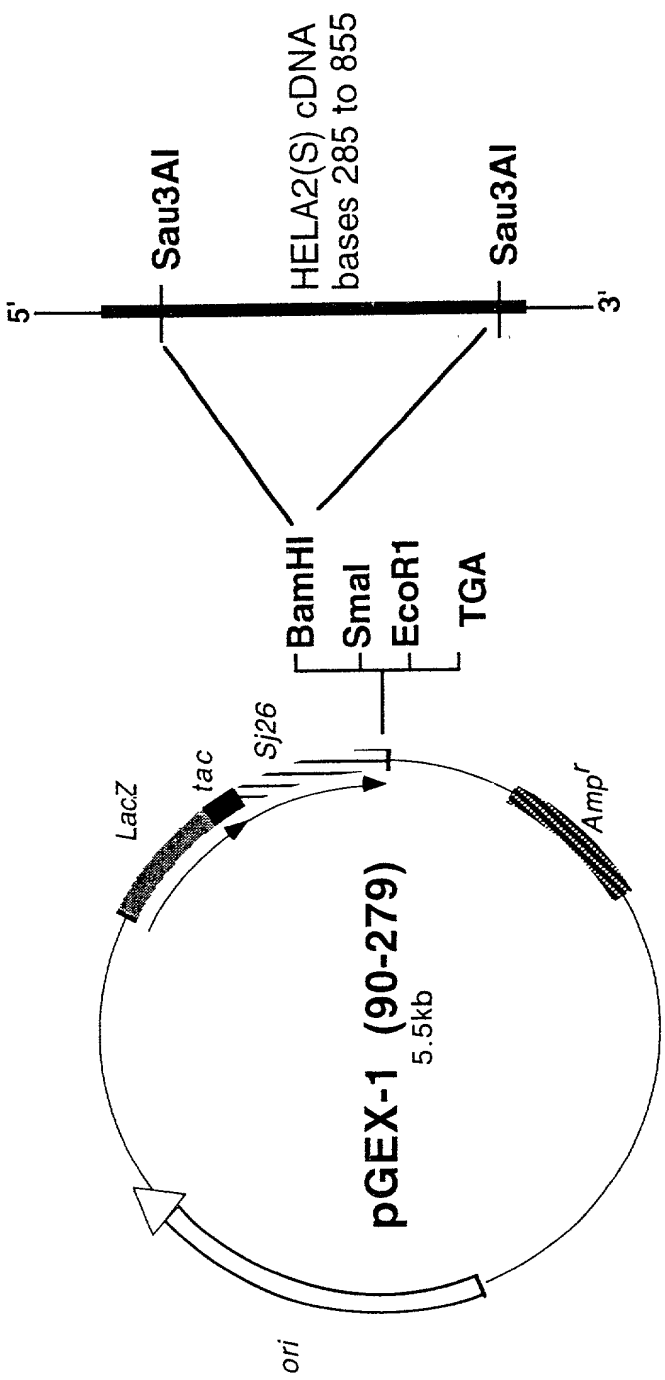
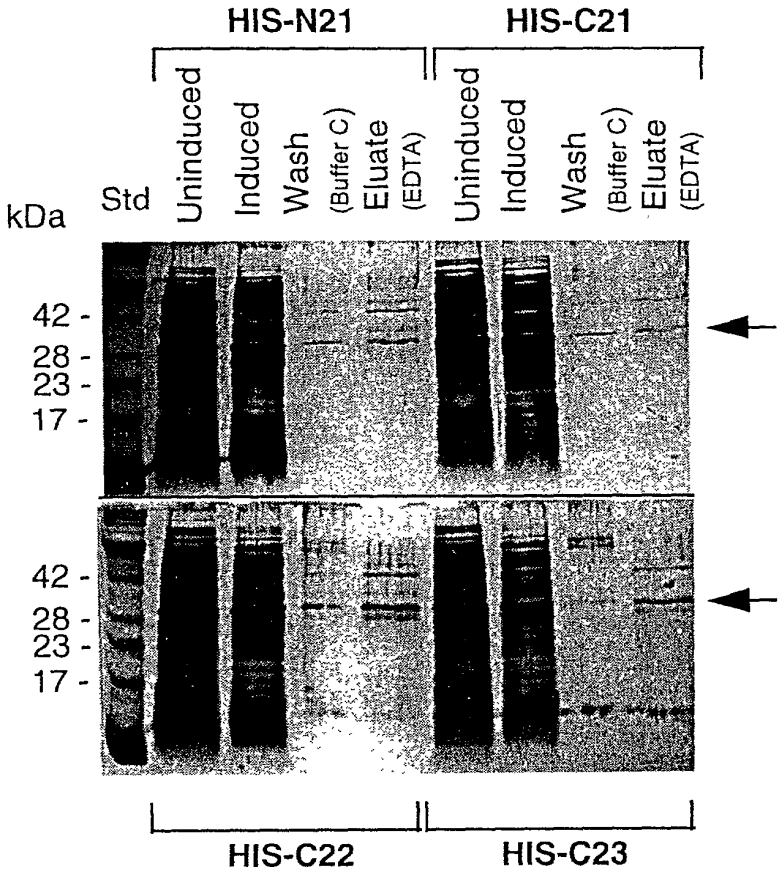

5'
Sau3AI
HELA2(S) cDNA
bases 285 to 855
Sau3AI
3'
BamHI
SmaI
EcoR1
TGA
Sj26
tac
LacZ
Amp^r
pGEX-1 (90-279)
5.5kb
ori

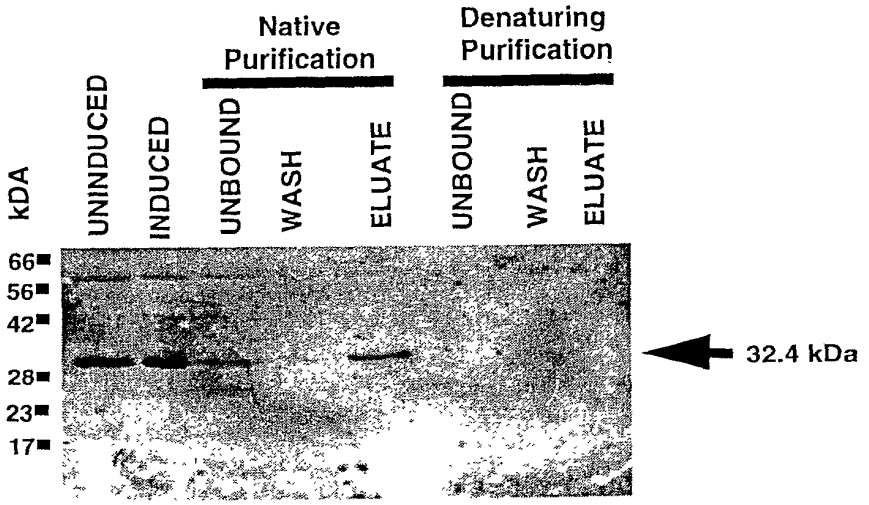
FIG 4(iii)

FIG 5

A. Expression of recombinant Testisin in *E. coli*.



B. Western blot of recombinant Testisin



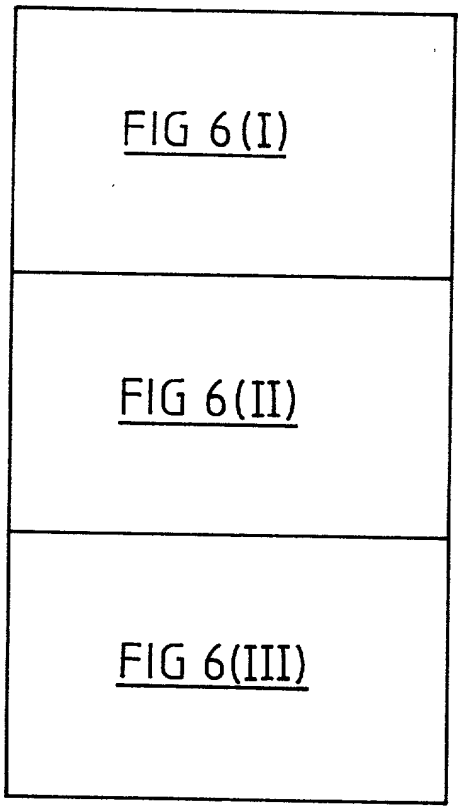


FIG 6

FIGURE 6 (I)

```
1                                                                GCCGCGGGAGAGGAGGCC
19   ATGGGCGCGCGCGGGGCGCTGCTGCTGGCGCTGCTGCTGGCTCGGGCTGGACTCAGGAAG
       M  G  A  R  G  A  L  L  L  A  L  L  L  A  R  A  G  L  R  K   20

79   CCGGAGTCGCAGGAGGCGGCGCCGTTATCAGGACCATGCGGCCGACGGGTCATCACGTCG
       P  E  S  Q  E  A  A  P  L  S  G  P  C  G  R  R  V  I  T  S   40

139  CGCATCGTGGGTGGAGAGGACGCCGAACTCGGGCGTTGGCCGTGGCAGGGGAGCCTGCGC
       R  I  V  G  G  E  D  A  E  L  G  R  W  P  W  Q  G  S  L  R   60

199  CTGTGGGATTCCCACGTATGCGGAGTGAGCCTGCTCAGCCACCGCTGGGCACTCACGGCG
       L  W  D  S  H  V  C  G  V  S  L  L  S  H  R  W  A  L  T  A   80

259  GCGCACTGCTTTGAAACCTATAGTGACCTTAGTGATCCCTCCGGGTGGATGGTCCAGTTT
       A  H  C  F  E  T  Y  S  D  L  S  D  P  S  G  W  M  V  Q  F  100

319  GGCCAGCTGACTTCCATGCCATCCTTCTGGAGCCTGCAGGCCTACTACACCCGTTACTTC
       G  Q  L  T  S  M  P  S  F  W  S  L  Q  A  Y  Y  T  R  Y  F  120

379  GTATCGAATATCTATCTGAGCCCTCGCTACCTGGGGAATTCACCCTATGACATTGCCTTG
       V  S  N  I  Y  L  S  P  R  Y  L  G  N  S  P  Y  D  I  A  L  140
```

FIGURE 6 (II)

```
439  GTGAAGCTGTCTGCACCTGTCACCTACACTAAACACATCCAGCCCATCTGTCTCCAGGCC
       V   K   L   S   A   P   V   T   Y   T   K   H   I   Q   P   I   C   L   Q   A  160

499  TCCACATTTGAGTTTGAGAACCGGACAGACTGCTGGGTGACTGGCTGGGGGTACATCAAA
       S   T   F   E   F   E   N   R   T   D   C   W   V   T   G   W   G   Y   I   K  180

559  GAGGATGAGGCACTGCCATCTCCCCACACCCTCCAGGAAGTTCAGGTCGCCATCATAAAC
       E   D   E   A   L   P   S   P   H   T   L   Q   E   V   Q   V   A   I   I   N  200

619  AACTCTATGTGCAACCACCTCTTCCTCAAGTACAGTTTCCGCAAGGACATCTTTGGAGAC
       N   S   M   C   N   H   L   F   L   K   Y   S   F   R   K   D   I   F   G   D  220

679  ATGGTTTGTGCTGGCAATGCCCAAGGCGGGAAGGATGCCTGCTTCGGTGACTCAGGTGGA
       M   V   C   A   G   N   A   Q   G   G   K   D   A   C   F   G   D   S   G   G  240

739  CCCTTGGCCTGTAACAAGAATGGACTGTGGTATCAGATTGGAGTCGTGAGCTGGGGAGTG
       P   L   A   C   N   K   N   G   L   W   Y   Q   I   G   V   V   S   W   G   V  260

799  GGCTGTGGTCGGCCCAATCGGCCCGGTGTCTACACCAATATCAGCCACCACTTTGAGTGG
       G   C   G   R   P   N   R   P   G   V   Y   T   N   I   S   H   H   F   E   W  280
```

FIGURE 6 (III)

```
859  ATCCAGAAGCTGATGGCCCAGAGTGGCATGTCCCAGCCAGACCCCTCCTGGCCGCTACTC
       I  Q  K  L  M  A  Q  S  G  M  S  Q  P  D  P  S  W  P  L  L  300

919  TTTTTCCCTCTTCTCTGGGCTCTCCCACTCCTGGGGCCGGTCTGAGCCTACCTGAGCCCA 314
       F  F  P  L  L  W  A  L  P  L  L  G  P  V  *

979  TGCAGCCTGGGGCCACTGCCAAGTCAGGCCCTGGTTCTCTTCTGTCTTGTTTGGTAATAA
1039 ACACATTCCAGTTGATGCCTTGCAGGGCATTCTTCAAAAAAAAAAAAAAAAAAAAAAAA
1099 AAAAAAAAAAAAAAAAAA
```

Western blot of GST-Testisin using anti-Testisin peptide T175 antibody

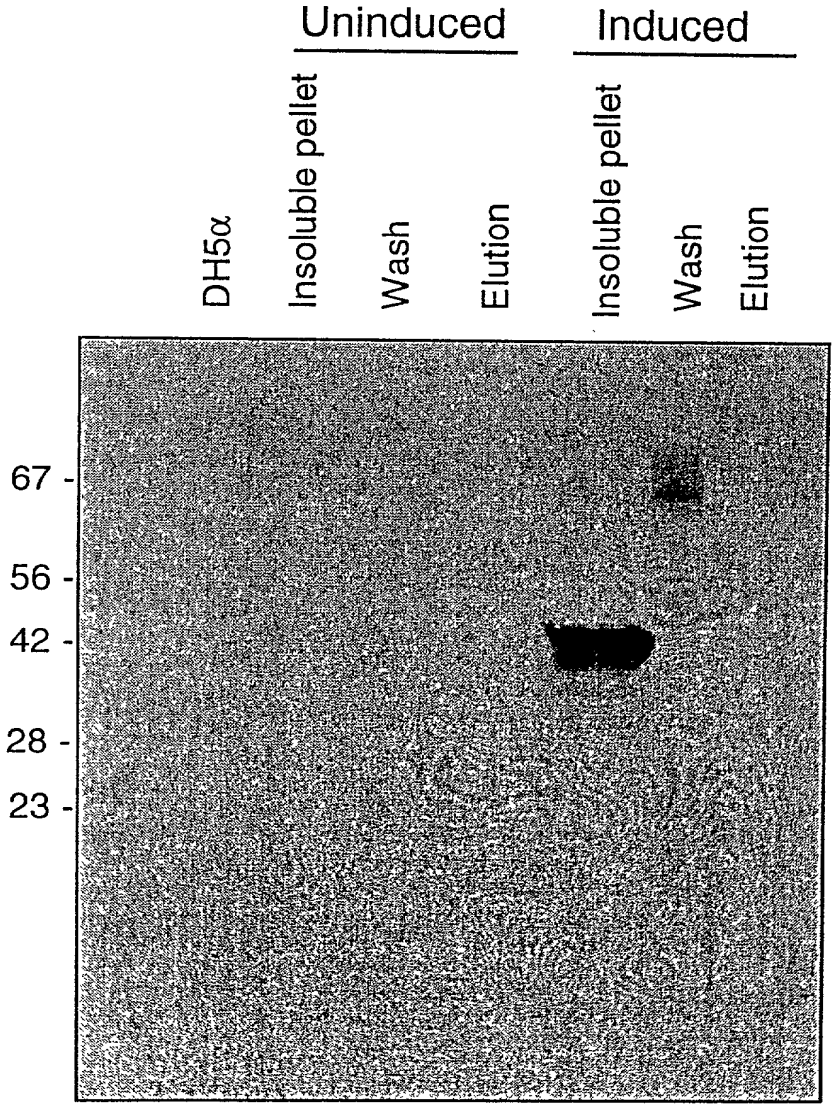
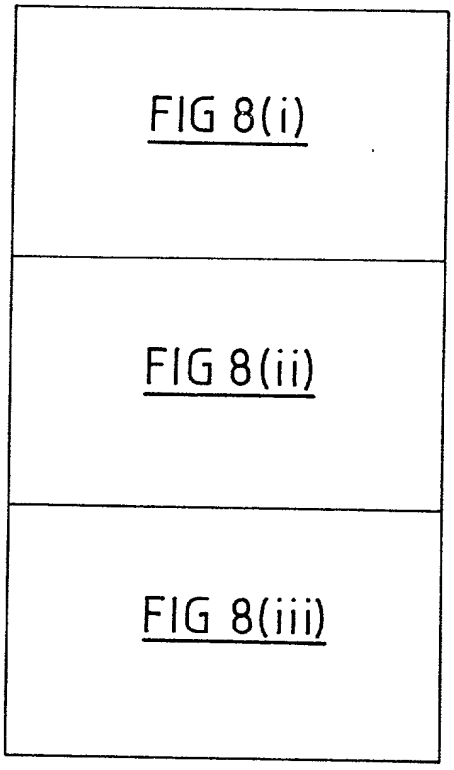


FIG 7

FIG 8



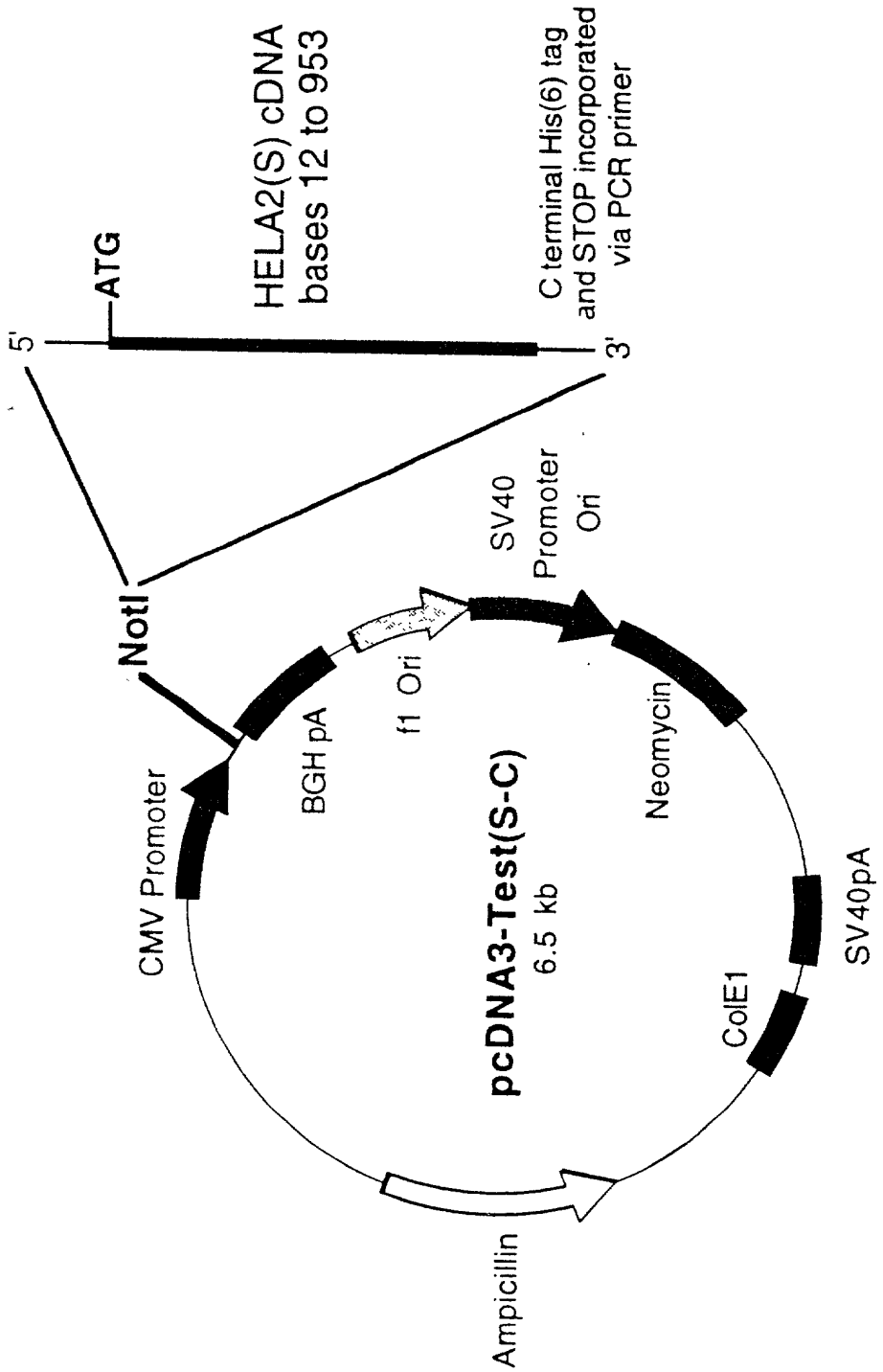

5'
ATG
HELA2(S) cDNA
bases 12 to 953
C terminal His(6) tag
and STOP incorporated
via PCR primer
3'
NotI
SV40
Promoter
Ori
CMV Promoter
BGH pA
f1 Ori
pcDNA3-Test(S-C)
6.5 kb
Neomycin
SV40pA
ColE1
Ampicillin

FIG 8(i)

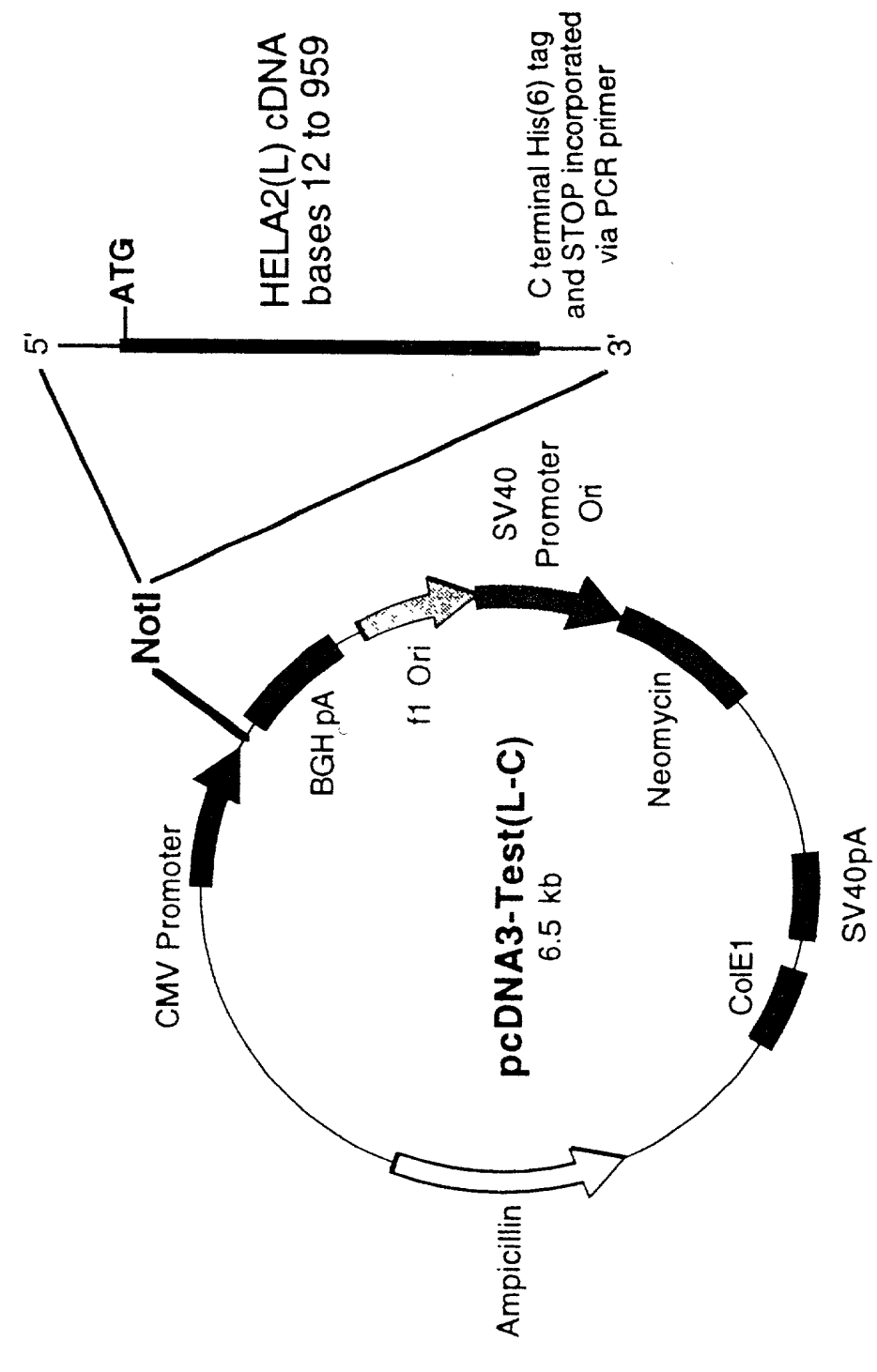

5'
ATG
HELA2(L) cDNA
bases 12 to 959
C terminal His(6) tag
and STOP incorporated
via PCR primer
3'
NotI
SV40
Promoter
Ori
f1 Ori
BGH pA
CMV Promoter
pcDNA3-Test(L-C)
6.5 kb
Neomycin
ColE1
SV40pA
Ampicillin

FIG 8(ii)

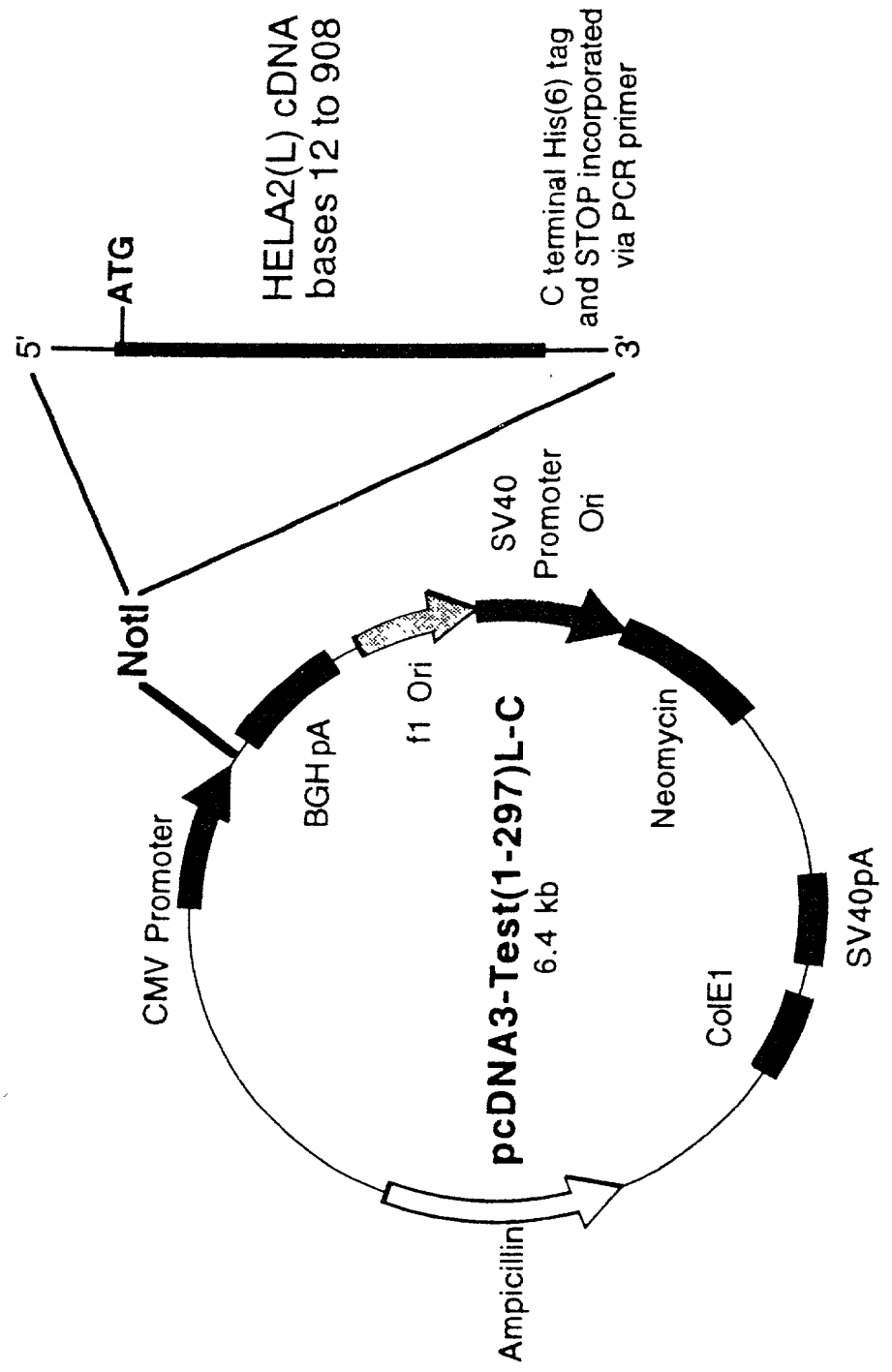

5'
ATG
HELA2(L) cDNA
bases 12 to 908
C terminal His(6) tag
and STOP incorporated
via PCR primer
3'
NotI
SV40
Promoter
Ori
f1 Ori
BGH pA
CMV Promoter
pcDNA3-Test(1-297)L-C
6.4 kb
Neomycin
SV40pA
ColE1
Ampicillin

FIG 8(iii)

FIG 9

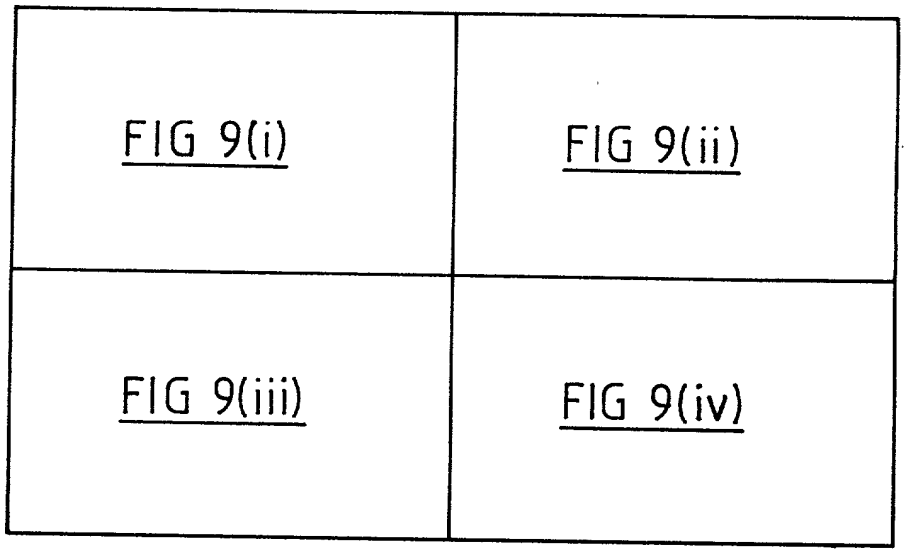
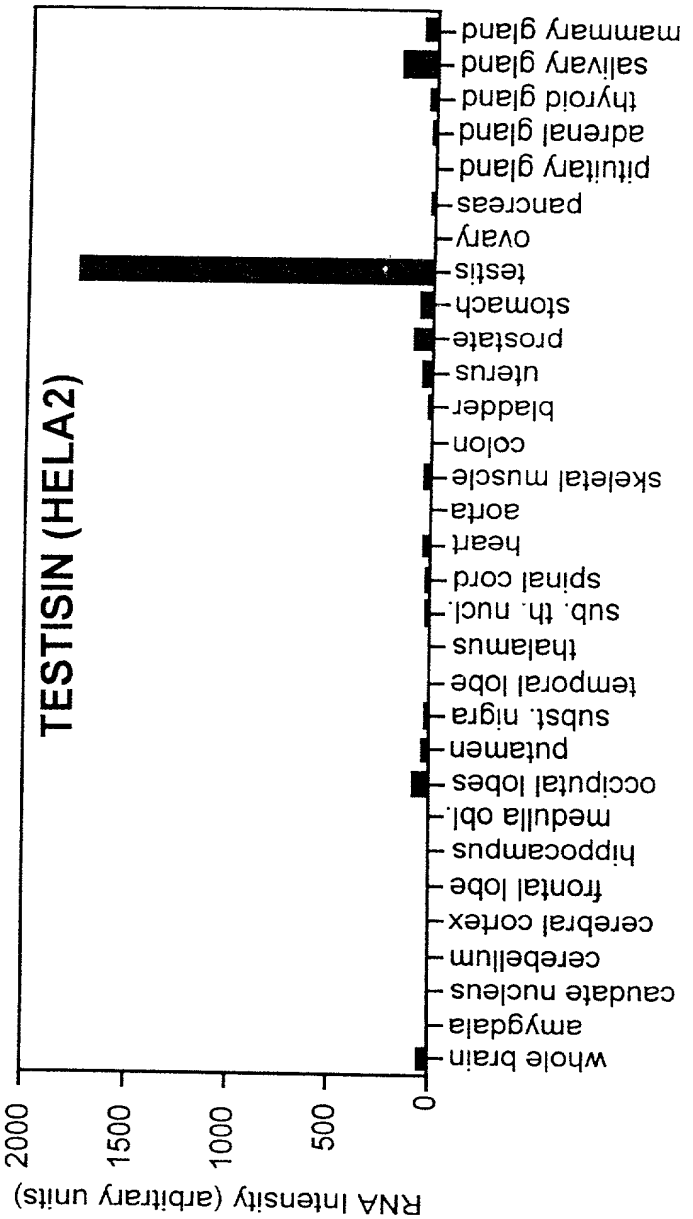


FIG 9(i)


TESTISIN (HELA2)
RNA intensity (arbitrary units)
0
500
1000
1500
2000
whole brain
amygdala
caudate nucleus
cerebellum
cerebral cortex
frontal lobe
hippocampus
medulla obl.
occiputal lobes
putamen
substr. nigra
temporal lobe
thalamus
sub. th. nucl.
spinal cord
heart
aorta
skeletal muscle
colon
bladder
uterus
prostate
stomach
testis
ovary
pancreas
pituitary gland
adrenal gland
thyroid gland
salivary gland
mammary gland

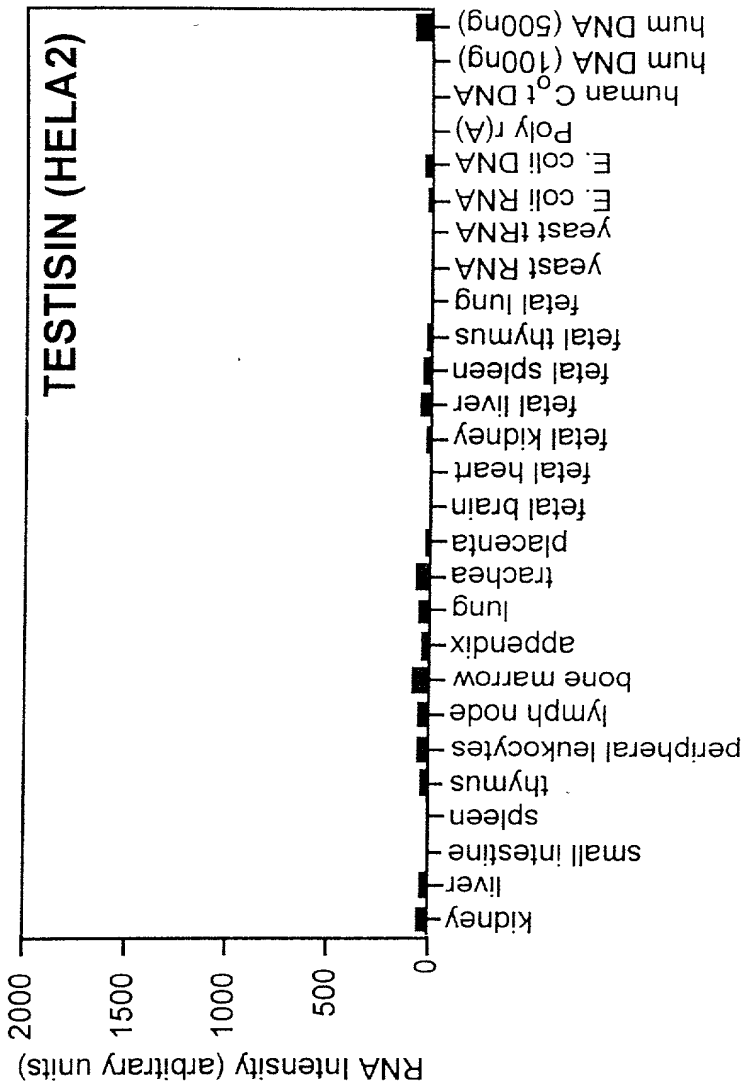

TESTISIN (HELA2)
RNA Intensity (arbitrary units)
0
500
1000
1500
2000
kidney
liver
small intestine
spleen
thymus
peripheral leukocytes
lymph node
bone marrow
appendix
lung
trachea
placenta
fetal brain
fetal heart
fetal kidney
fetal liver
fetal spleen
fetal thymus
fetal lung
yeast RNA
yeast tRNA
E. coli RNA
E. coli DNA
Poly r(A)
human $C_o t$ DNA
hum DNA (100ng)
hum DNA (500ng)

FIG 9(ii)

FIG 9(iii)

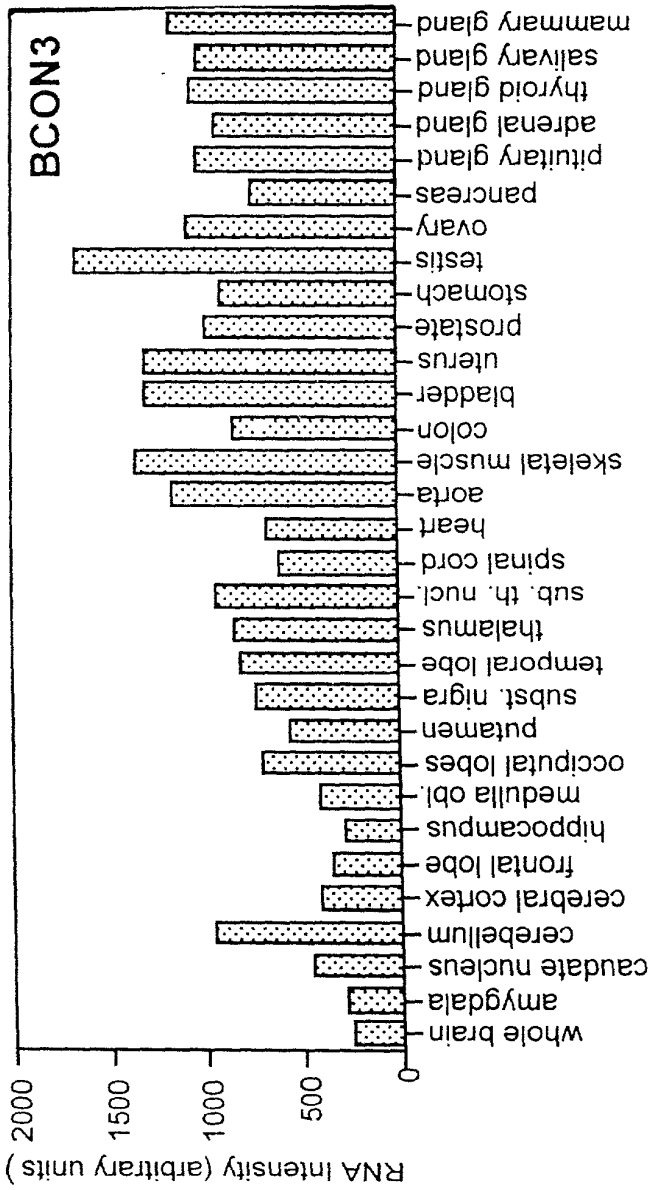
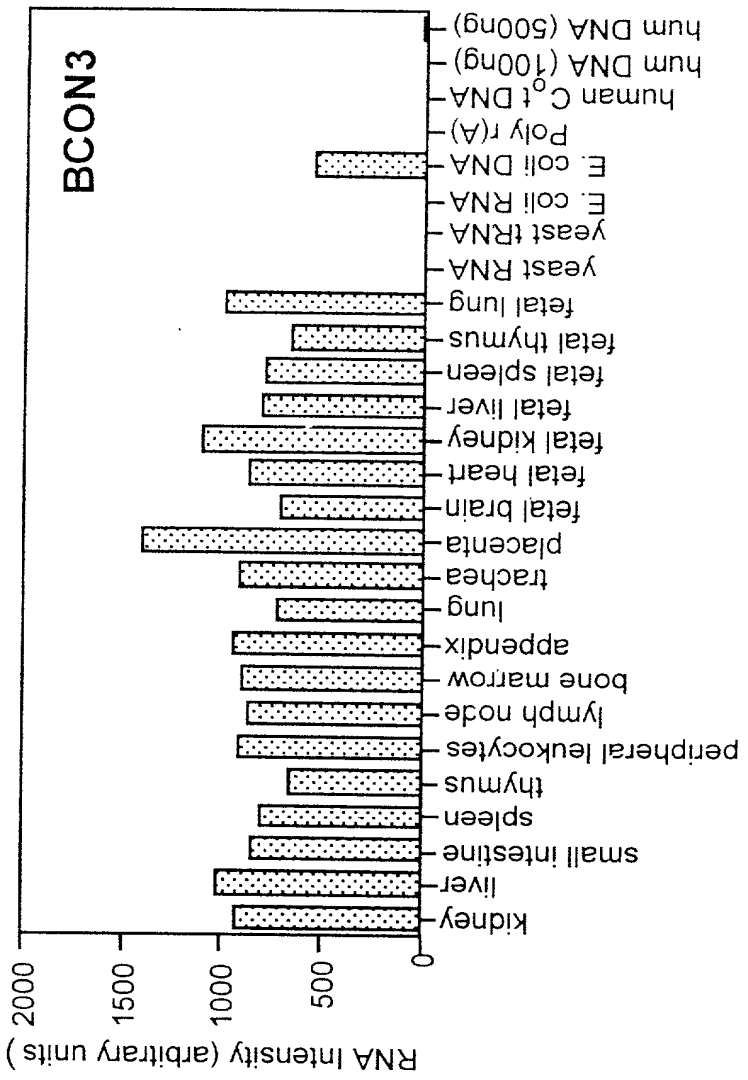

BCON3
RNA intensity (arbitrary units)
0
500
1000
1500
2000
whole brain
amygdala
caudate nucleus
cerebellum
cerebral cortex
frontal lobe
hippocampus
medulla obl.
occiputal lobes
putamen
substr. nigra
temporal lobe
thalamus
sub. th. nucl.
spinal cord
heart
aorta
skeletal muscle
colon
bladder
uterus
prostate
stomach
testis
ovary
pancreas
pituitary gland
adrenal gland
thyroid gland
salivary gland
mammary gland

FIG 9(iv)


BCON3
RNA Intensity (arbitrary units)
2000
1500
1000
500
0
kidney
liver
small intestine
spleen
thymus
peripheral leukocytes
lymph node
bone marrow
appendix
lung
trachea
placenta
fetal brain
fetal heart
fetal kidney
fetal liver
fetal spleen
fetal thymus
fetal lung
yeast RNA
yeast tRNA
E. coli RNA
E. coli DNA
Poly r(A)
human C$_o$t DNA
hum DNA (100ng)
hum DNA (500ng)

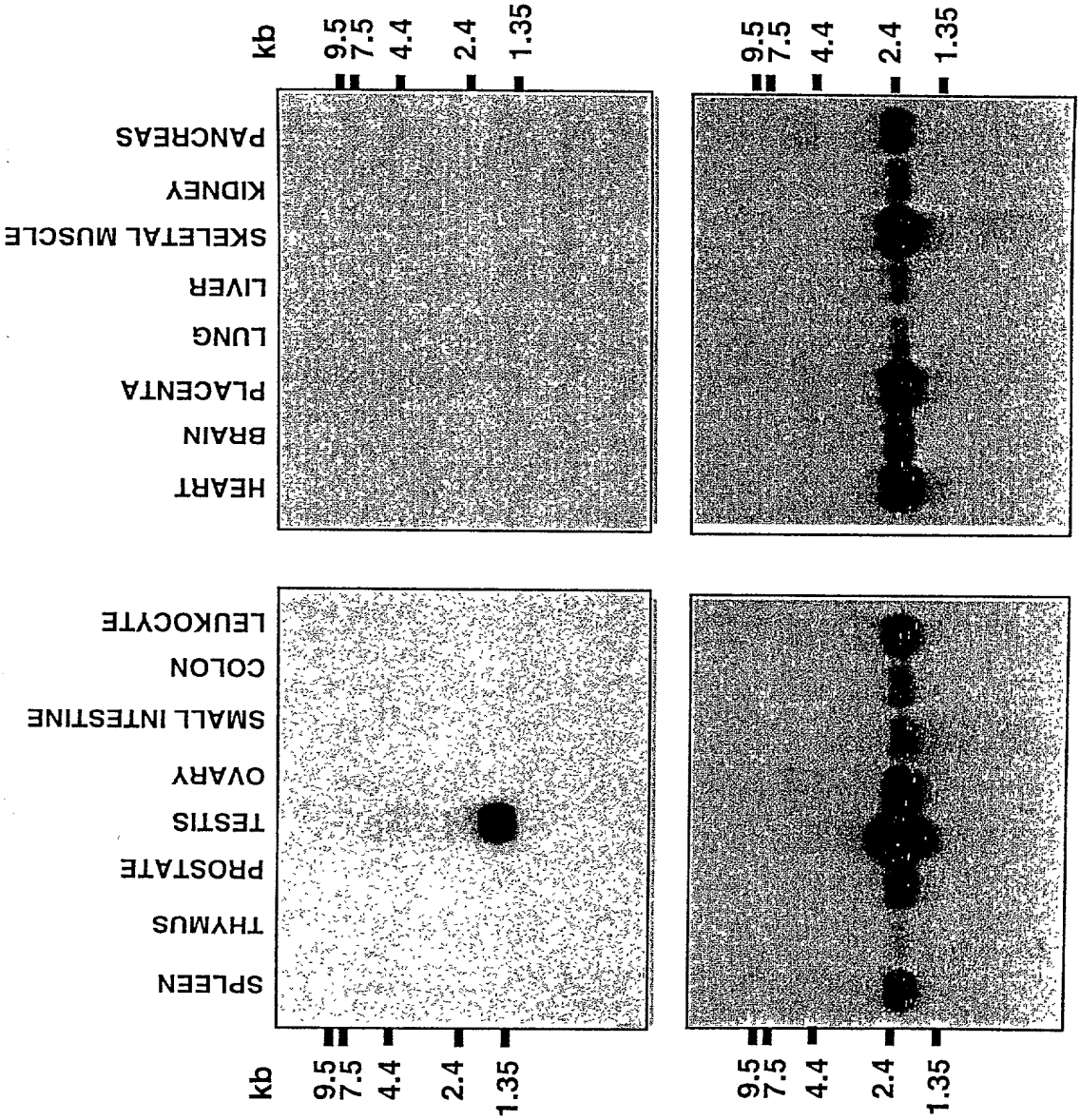
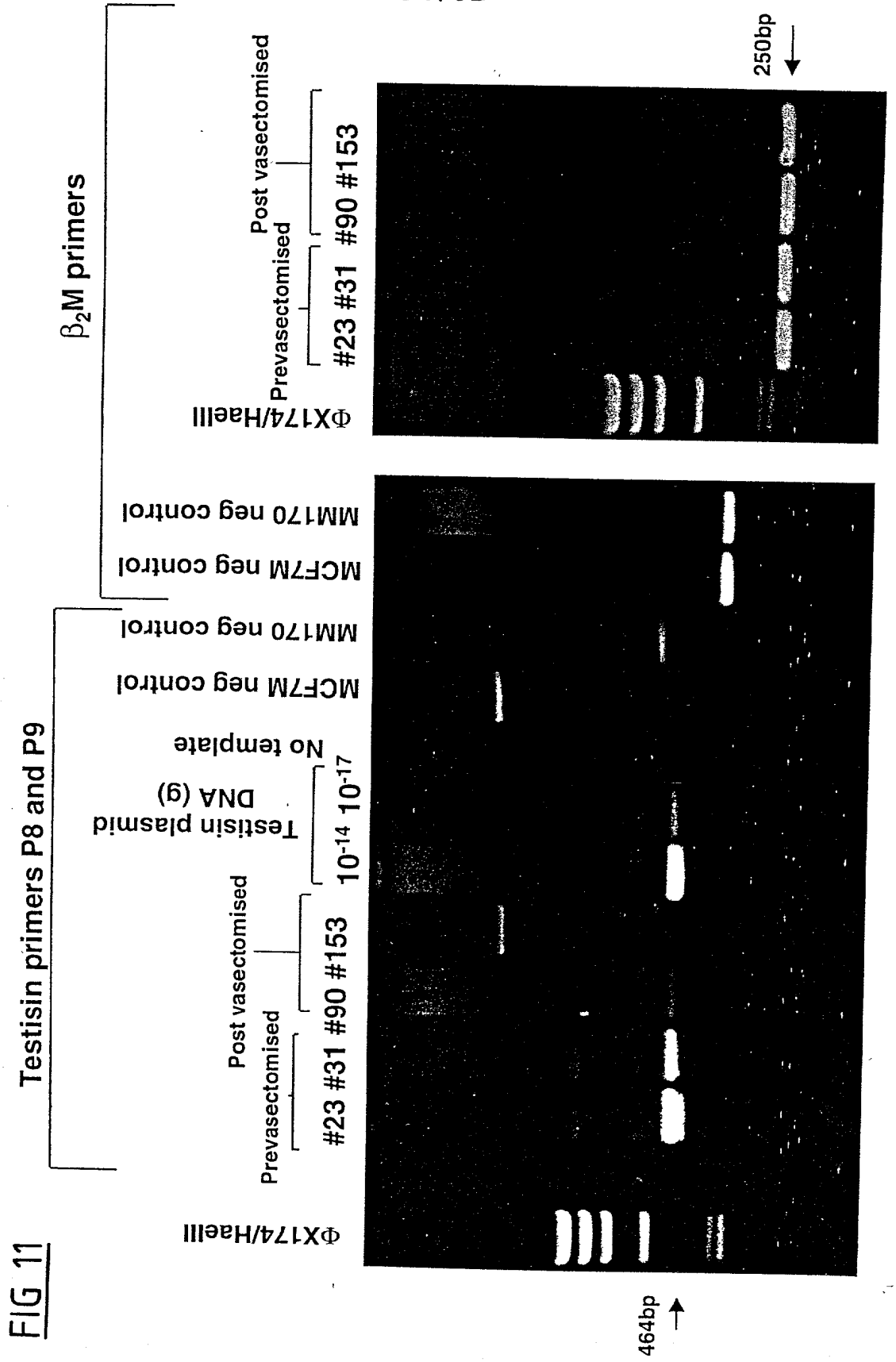

A.
HELA2 (Testisin)
B.
BCON3
kb
9.5
7.5
4.4
2.4
1.35
SPLEEN
THYMUS
PROSTATE
TESTIS
OVARY
SMALL INTESTINE
COLON
LEUKOCYTE
HEART
BRAIN
PLACENTA
LUNG
LIVER
SKELETAL MUSCLE
KIDNEY
PANCREAS

FIG 10

FIG 11


Testisin primers P8 and P9
β2M primers
ΦX174/HaeIII
Prevasectomised
Post vasectomised
#23 #31 #90 #153
Testisin plasmid DNA (g)
10-14 10-17
No template
MCF7M neg control
MM170 neg control
MCF7M neg control
MM170 neg control
ΦX174/HaeIII
Prevasectomised
Post vasectomised
#23 #31 #90 #153
464bp
250bp

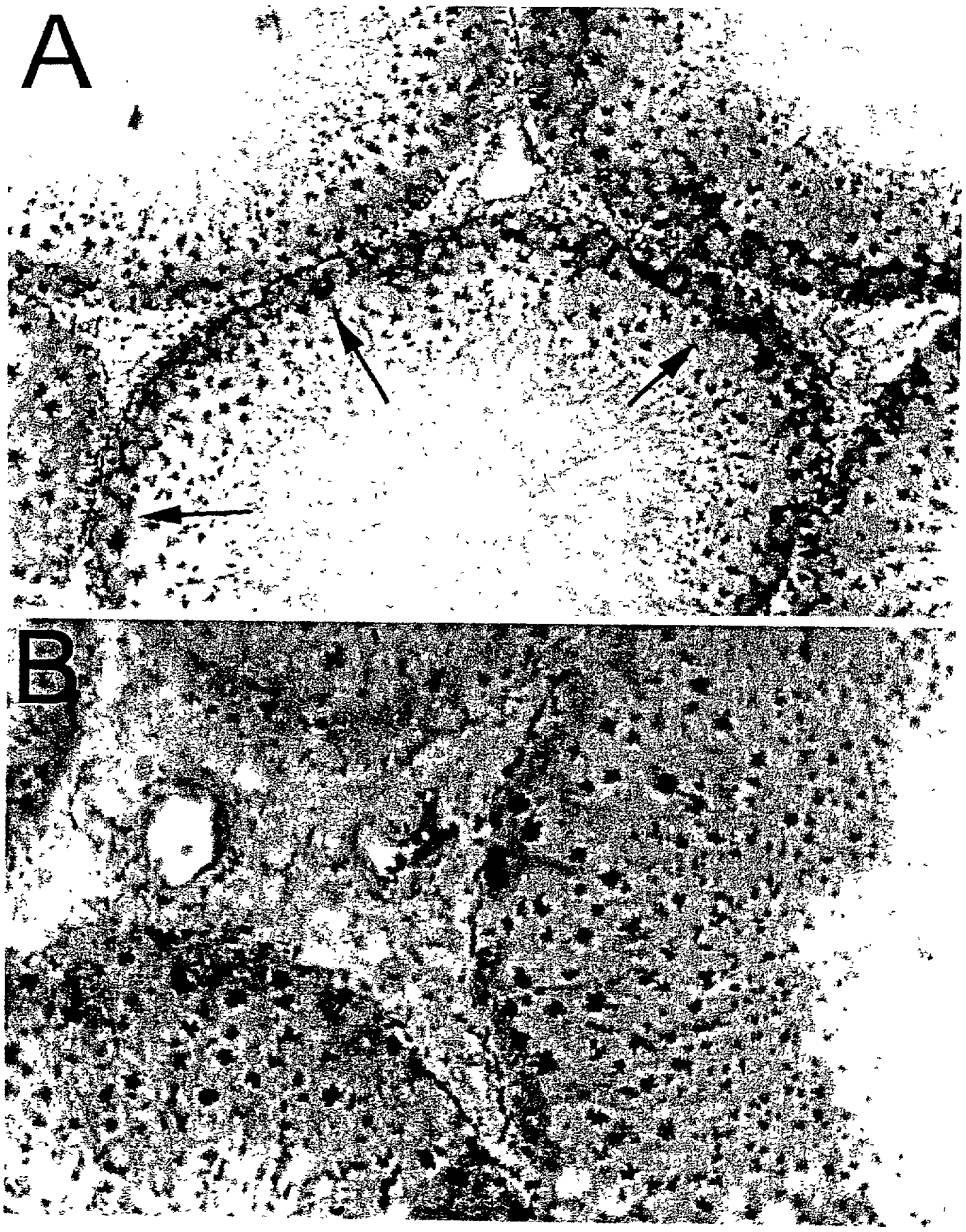


FIG 12

Testisin (HELA2) is located on human chromosome 16p13.3

A

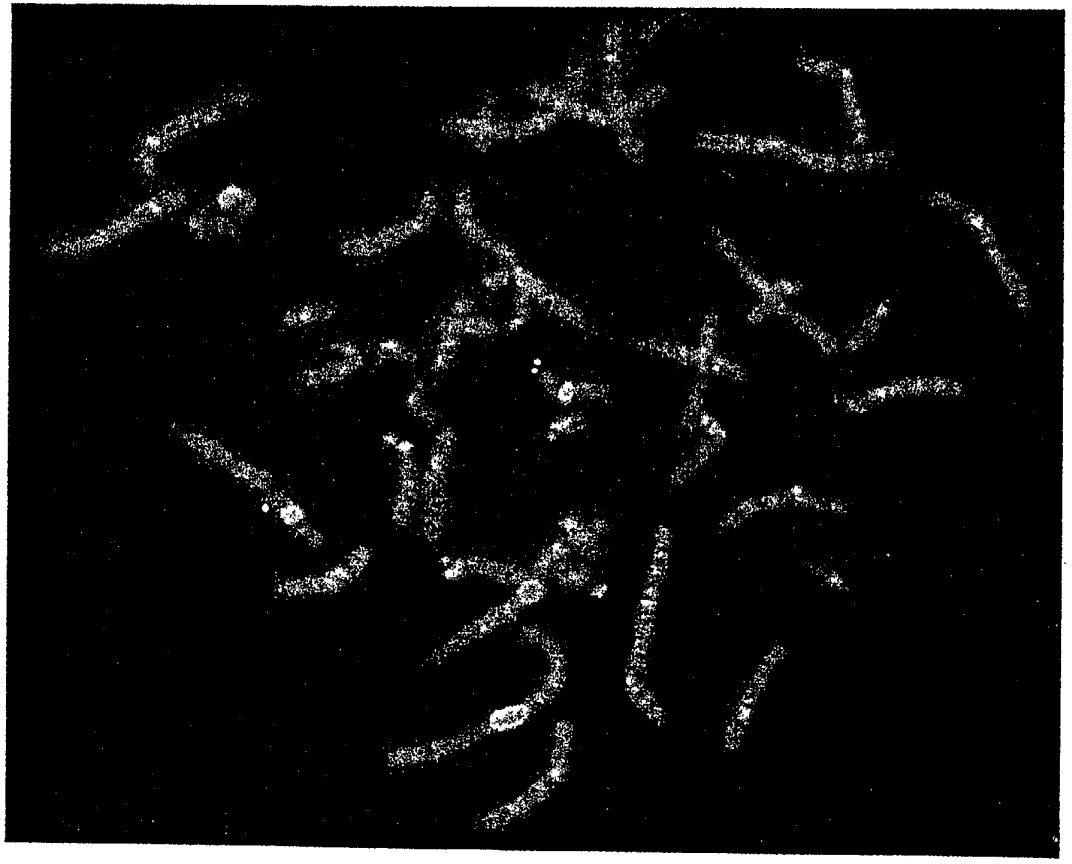


FIG 13A

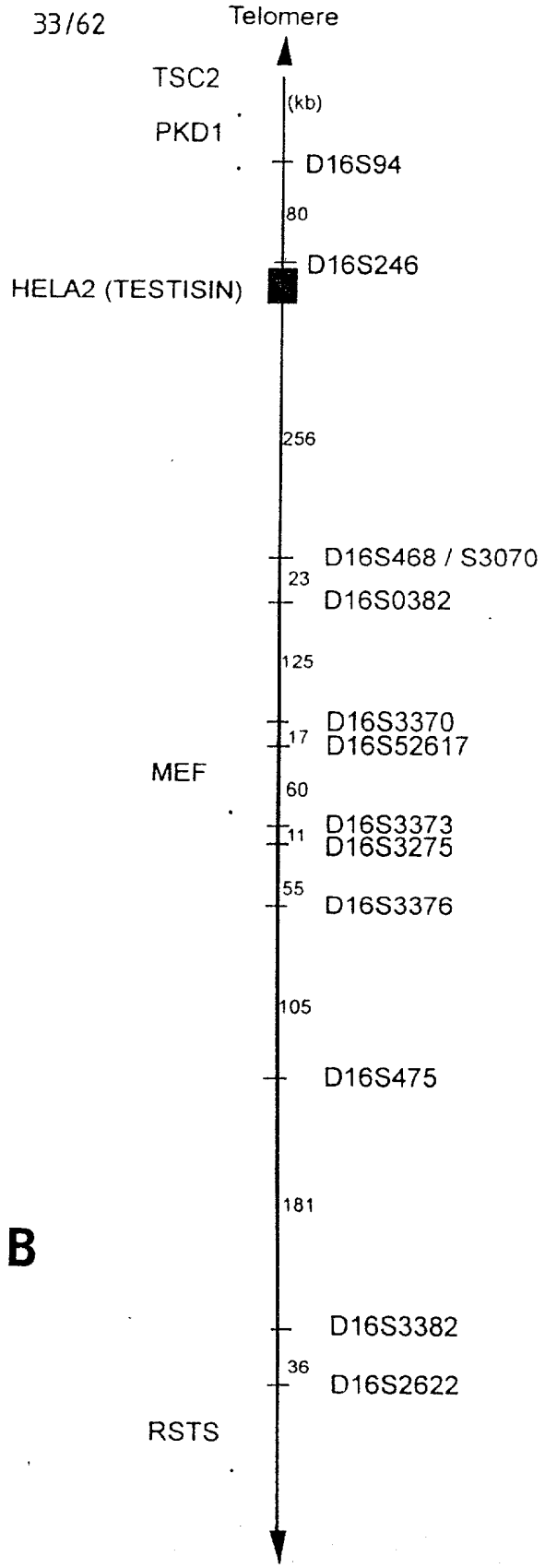
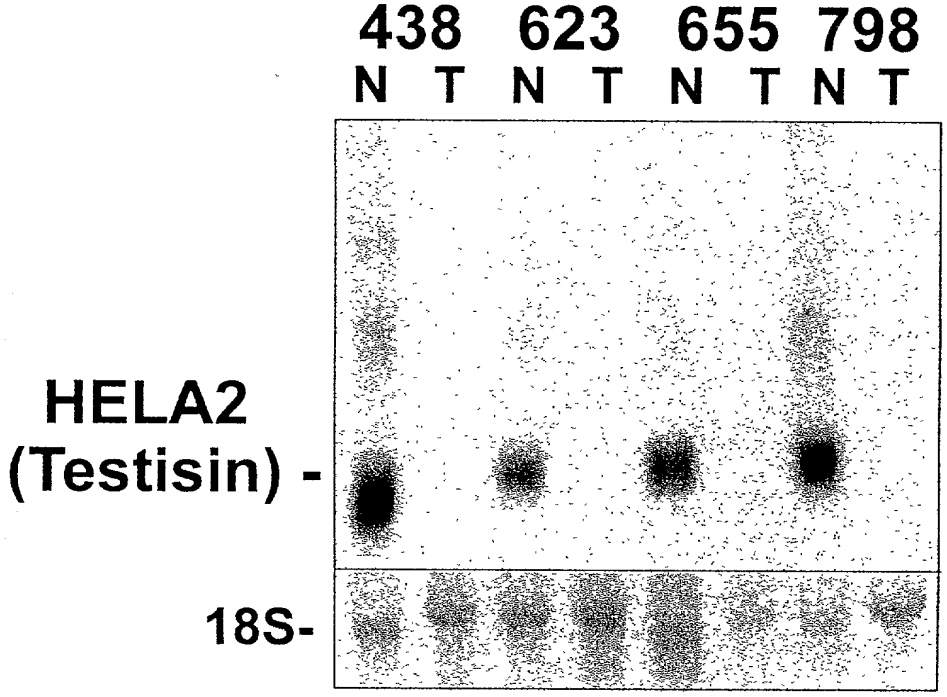

Telomere
TSC2
(kb)
PKD1
D16S94
80
D16S246
HELA2 (TESTISIN)
256
D16S468 / S3070
23
D16S0382
125
D16S3370
17
D16S52617
MEF
60
D16S3373
11
D16S3275
55
D16S3376
105
D16S475
181
D16S3382
36
D16S2622
RSTS

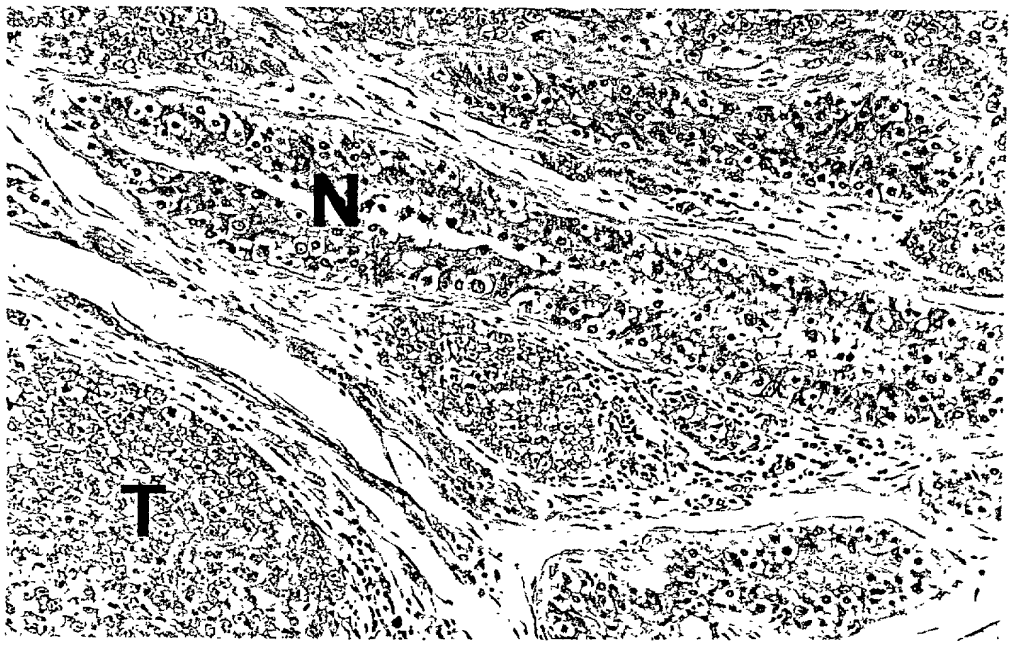
FIGURE 13 B

FIG 14 34/62

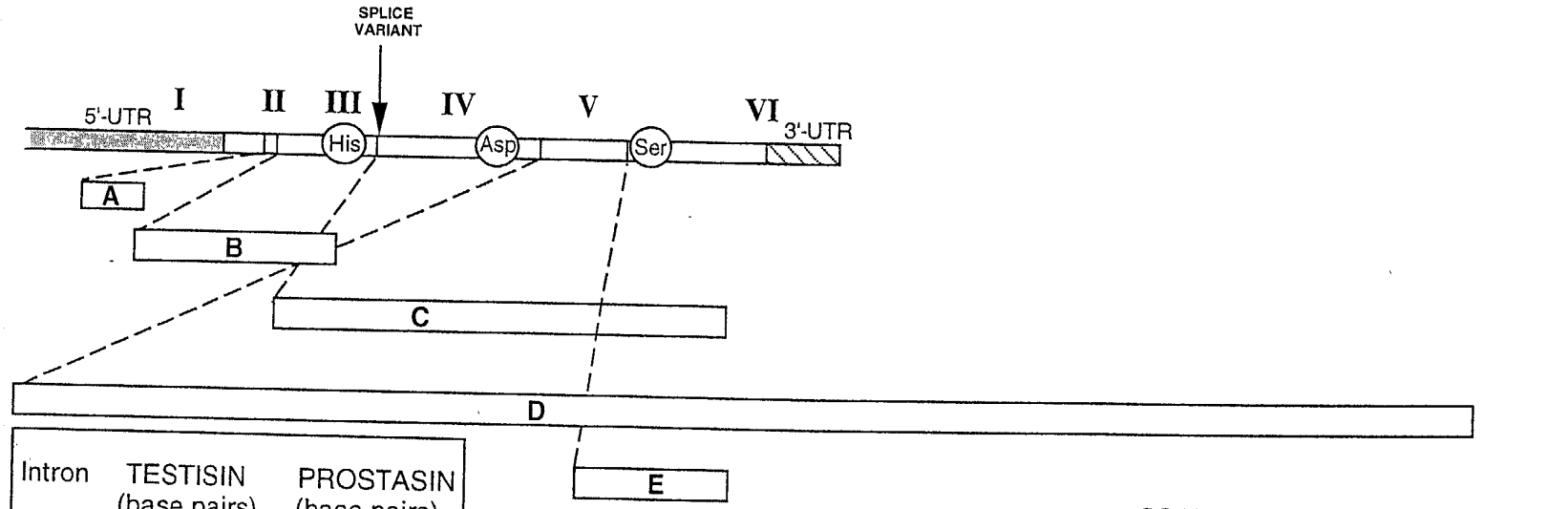
A. Northern Blot



B. Immunohistochemistry



TESTISIN INTRON/EXON BOUNDARIES AND SIZES



Intron	TESTISIN (base pairs)	PROSTASIN (base pairs)
A	97	243
B	344	1763
C	716	271
D	~2200	85
E	256	92

Exon	TESTISIN (base pairs)	PROSTASIN (base pairs)
I	>76	417
II	18	18
III	163	163
IV	284	272
V	168	167
VI	348	899

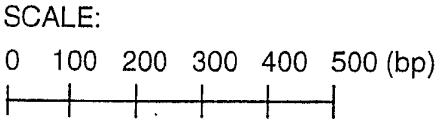
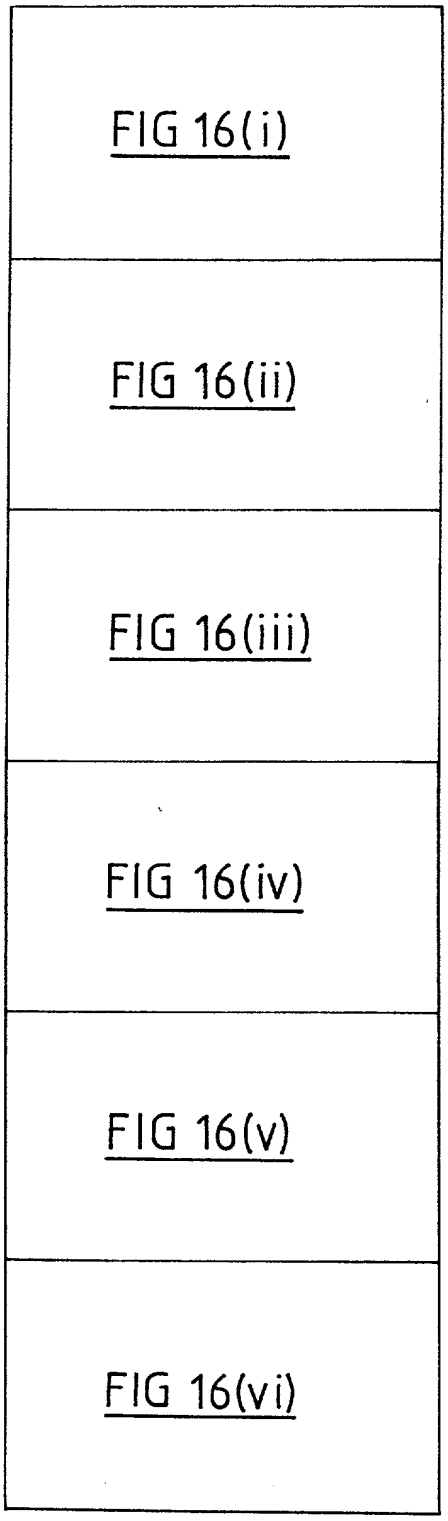


FIGURE 15

FIG 16



```
agtgagtctc ctgcctcagc ctcccaagta gctgggactt caggtgtgtg     50
ccaccatcct cagctaattt tttttttttt tttttttttg agaaggagtc    100
ttgctctgtc gcccaggctg gagtgcagtg gcgcgatctt ccaggcccca    150
ccgggccctc aggaaggcct tgcctacctg ctttaagggg actcctggct    200
cagggccagg cccctggtgc tggaggaggt ggtgggtgga gggcaggggg    250
caccaagcgg gcagccagga cccccgggct gcagacaaga aaaggactgt    300
                     /+1...EXON 1...
ggggtccacc gggtctgggc cACATCAAGG AATGTGGTTG AAGACCCGCC    350
CTTAGGAGCT GAAAGCCAGG GCGCTACCAG GCCTGAGAGG CCCCAAACAG    400
CCCTTGGGCC TGGTTTGGGA GGATTAAGCT GGAGCTCCCA ACCCGCCCTG    450
CCCCCAGGGG GCGACCCCGG GCCCGGCGCG AGAGGAGGCA GAGGGGGCGT    500
CAGGCCGCGG GAGAGGAGGC CATGGGCGCG CGCGGGGCGC TGCTGCTGGC    550
                                        /INTRON A...
GCTGCTGCTG GCTCGGGCTG GACTCAGGAA GCCGGgtgag ctcggggcgc    600
tgctggcggg atggggaggc gggggagcgg tggggaggac gggaggtgga    650
```

FIG 16(i)

/EXON 2...

ggccgcgggg agtcacttct tgtctcccgc agAGTCGCAG GAGGCGGCGC 700

/INTRON B...

CGTTATCAGg tagggcgccc aggacgcgcg attcctgcca gggccgttgg 750

gccgaggtgg acgggggggcg gtgaggggggt agaggggggc ctttactgct 800

ctctcgcccc cgccccccggg atcgagaact ctgttggcgt ggaaagtaac 850

taacggacgc tggaggggga tgggcgggcc ctgcagagca cgtgggagga 900

tctccagtgt cacctacttc ctgctgcaca cacgcgaggg gaccctgggt 950

gggcaaaaac gtgctttccc ggacggggtt gaaggggaga aagggagagg 1000

tcgggcttgg ggggctgcct cccgcggctc agcagttcct ctgaccatcc 1050

/EXON 3...

gagGACCATG CGGCCGACGG GTCATCACGT CGCGCATCGT GGGTGGAGAG 1100

GACGCCGAAC TCGGGCGTTG GCCGTGGCAG GGGAGCCTGC GCCTGTGGGA 1150

TTCCCACGTA TGCGGAGTGA GCCTGCTCAG CCACCGCTGG GCACTCACGG 1200

FIG 16(ii)

/INTRON C...

```
CGGCGCACTG CTTTGAAACg tgagtggggg tgcgaacgga ggggtgcggg   1250
gacgggcagg aacagggctg gagggagtgc caccgaactt tacctctggt   1300
ctgatgccag acttgggcgt gaaagttgtg cgtggatgcg gcctggtgtt   1350
ctcctgagcc ccaggctgtg ctgcagccgg ttacacccac tccagttccc   1400
tttgggtctc ctggagggaa ccctgttcag gttattccag aatgttcttc   1450
cagaacattt ccacacactt ttgggtattc tctccctttt tctttcaacc   1500
caaagttcac cactgaccat cccaccctca tccccccctcc tggtggacgg   1550
tgcggtacag tgtggggcac tgagccaagg ccagcacccc cgggccgctg   1600
tgtggactcc atcctgccaa tcccacattg gcgtggtgca tctccccatt   1650
cctccttggg ctgcatgggg gtgcccctgg aggccttggc tcaatgcaag   1700
gctccttggg acagctctgg gaggtgacaa gaccccaccc ttctgctgca   1750
ggagcaggtc ctaggacttt ggttgtggtc tgtctgggct ccttcatttc   1800
tgcaggggac cctgggtgtt agcaagtagc agcaacacca cagtttcccc   1850
tcctgcactg gaccccagtt gtgctcaggt agccagccct ccatccaggg   1900
```

FIG 16(iii)

/EXON 4...

```
cccctgactg ctctcttctc ttctgccagc tatagTGACC TTAGTGATCC      1950
CTCCGGGTGG ATGGTCCAGT TTGGCCAGCT GACTTCCATG CCATCCTTCT      2000
GGAGCCTGCA GGCCTACTAC ACCCGTTACT TCGTATCGAA TATCTATCTG      2050
AGCCCTCGCT ACCTGGGGAA TTCACCCTAT GACATTGCCT TGGTGAAGCT      2100
GTCTGCACCT GTCACCTACA CTAAACACAT CCAGCCCATC TGTCTCCAGG      2150
CCTCCACATT TGAGTTTGAG AACCGGACAG ACTGCTGGGT GACTGGCTGG      2200
                        /INTRON  D...
GGGTACATCA AAGAGGATGA GGgtgaggct ggggacaggc gggtcaggga      2250
ggaactgtct ttgttcacct gttcccctgc ataggcacaa tagccccctg      2300
cttggtctgg gggtgcaggc tatgcccctc ttgcttgcag tctctcctca      2350
cctgccaggg cagggaccaa acacccagtt ctctcccttc caggggctgt      2400
gggggccaga aggagagtgt gagagggagg ccagtttggc gcaagcctgt      2450
gggtggtgcg gtggtggagg ggttctggag ggcttggcga cataaacctc      2500
atacttggat ttattcctgc atctttccac ctcccccagt gctcaccaat      2550
```

FIG 16(iv)

```
gccccaggca tca..............approx 1000 bp...........            3563
ccaggttgcc ccttccccca aggtctggct ttggatgctt atgtgaacac          ≈3613
cgttttaagt tgccttggcc ccttcctcgg ttcctttttg gctgaggaat          ≈3663
ctctccatgg ctgcaggcag ggccattgtt gccattctac agatagggaa          ≈3713
agtgcggctg ggggagctct gacagctgtc cctccccggg gccttctgtg          ≈3763
atgctgctga gggcctctgt tgtgctgggg tctgggttgg agctgggggt          ≈3813
aatggagatg aacctgccag gcacagtggg tgccccaggg cccccacccc          ≈3863
cgcagcctat gccatccctc catagagggg cctcaggttg ctgtctctct          ≈3913
                        /EXON 5...
ccttcccact atcgtccgca cagCACTGCC ATCTCCCCAC ACCCTCCAGG          ≈3963
AAGTTCAGGT CGCCATCATA AACAACTCTA TGTGCAACCA CCTCTTCCTC          ≈4013
AAGTACAGTT TCCGCAAGGA CATCTTTGGA GACATGGTTT GTGCTGGCAA          ≈4063
                              /INTRON  E...
TGCCCAAGGC GGGAAGGATG CCTGCTTCgt gagtgtcctt gccaccactc          ≈4113
ccagcccagg aaagcatcct gtgtccctgt gccttatttg accctcatgc          ≈4163
caaccccggg aggtggagac tgttgcccca ctctgcagat gcagaaacgg          ≈4213
```

FIG 16(v)

```
aggcttggct gctgccaggg ggaggaggag gatgtgcacc cagtctaccc      ≈4263
agccccatag cccttcccac tctcagcccc tcccctgccc cactcactct      ≈4313
                                        /EXON 6...
gccccaggct gacctcagcc ccgctgctcc ccagGGTGAC TCAGGTGGAC      ≈4363
CCTTGGCCTG TAACAAGAAT GGACTGTGGT ATCAGATTGG AGTCGTGAGC      ≈4413
TGGGGAGTGG GCTGTGGTCG GCCCAATCGG CCCGGTGTCT ACACCAATAT      ≈4463
CAGCCACCAC TTTGAGTGGA TCCAGAAGCT GATGGCCCAG AGTGGCATGT      ≈4513
CCCAGCCAGA CCCCTCCTGG CCGCTACTCT TTTTCCCTCT TCTCTGGGCT      ≈4563
CTCCCACTCC TGGGGCCGGT CTGAGCCTAC CTGAGCCCAT GCAGCCTGGG      ≈4613
GCCACTGCCA AGTCAGGCCC TGGTTCTCTT CTGTCTTGTT TGGTAATAAA      ≈4663
CACATTCCAG TTGATGCCTT GCAGGGCATT CTTCAaaagc agtggcttca      ≈4713
tggacagctc attctctctt gtgcagacag cctgtctgtg cccctggctc      ≈4763
acacccacat ctgttctgca ccatagaacc atctggttat ttcgatcaga      ≈4813
aagagaattg tgtgttgccc aggctggtct tgaacgccta gggtgtctcg      ≈4863
atc                                                         ≈4866
```

FIG 16(vi)

EXON III CACTGCTTTGAAAC**gt**gagtgggggtgcgaacggag
gggtgcggggacgggcaggaacagggctggagggagtgccaccga
actttacctctggtctgatgccagacttgggcgtgaaagttgtgc
gtggatgcggcctggtgttctcctgagccccaggctgtgctgcag
ccggttacacccactccagttccctttgggtctcctggagggaac
cctgttcaggttattccagaatgttcttccagaacatttccacac
acttttgggtattctctccctttttctttcaacccaaagttcacc
actgaccatcccaccctcatcccccctcctggtggacggtgcggt
acagtgtggggcactgagccaaggccagcacccccgggccgctgt

.....INTRON C (716 BP).....

gtggactccatcctgccaatcccacattggcgtggtgcatctccc
cattcctccttgggctgcatgggggtgcccctggaggccttggct
caatgcaaggctccttgggacagctctgggaggtgacaagacccc
acccttctgctgcaggagcaggtcctagactttggttgtggtctg
tctgggctccttcatttctgcaggggaccctgggtgttagcaagt
agcagcaacaccacagtttcccctcctgcactggaccccagttgt
gctcaggtagccagccctccatccagggcccctgactgctctctt
ctcttctgcc**ag**ctat**ag**TGACCTTAGTGATCCC EXON IV

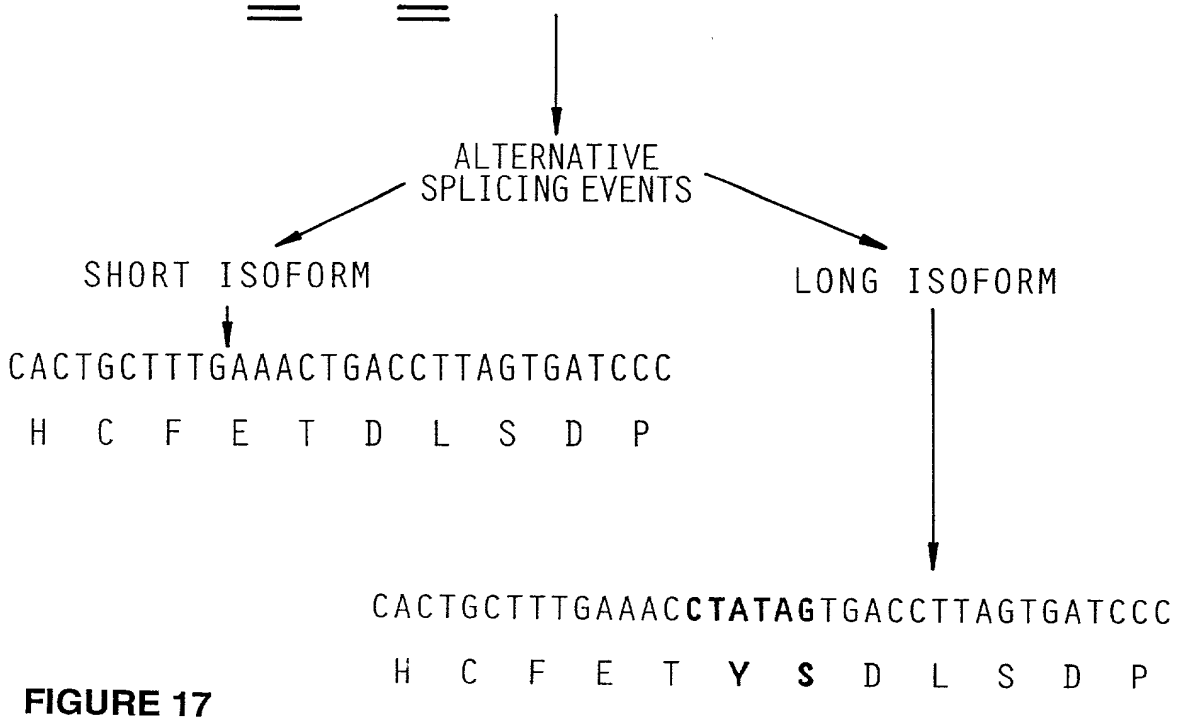


FIGURE 17

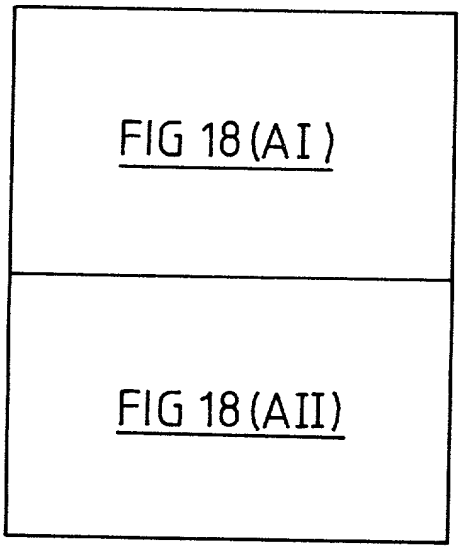


FIG 18(A)

FIGURE 18 (AI)

```
1   CGACCTATTGTCAGGGCCCTGCGGTCACAGGACCATCCCTTCCCGTATAGTGGGTGGCGA
     D  L  L  S  G  P  C  G  H  R  T  I  P  S  R  I  V  G  G  D  20

61  TGATGCTGAGCTTGGCCGCTGGCCGTGGCAAGGGAGCCTGCGTGTATGGGGCAACCACTT
     D  A  E  L  G  R  W  P  W  Q  G  S  L  R  V  W  G  N  H  L  40

121 ATGTGGCGCAACCTTGCTCAACCGCCGCTGGGTGCTTACAGCTGCCCACTGCTTCCAAAA
     C  G  A  T  L  L  N  R  R  W  V  L  T  A  A  H  C  F  Q  K  60

181 GGATAACGATCCTTTTGACTGGACAGTCCAGTTTGGTGAGCTGACTTCCAGGCCATCTCT
     D  N  D  P  F  D  W  T  V  Q  F  G  E  L  T  S  R  P  S  L  80

241 CTGGAACCTACAGGCCTATTCCAACCGTTACCAAATAGAAGATATTTTCCTGAGCCCCAA
     W  N  L  Q  A  Y  S  N  R  Y  Q  I  E  D  I  F  L  S  P  K 100

301 GTACTCGGAGCAGTATCCCAATGACATAGCCCTGCTGAAGCTGTCATCTCCAGTCACCTA
     Y  S  E  Q  Y  P  N  D  I  A  L  L  K  L  S  S  P  V  T  Y 120

361 CAATAACTTCATCCAGCCCATCTGCCTCCTGAACTCCACGTACAAGTTTGAGAACCGAAC
     N  N  F  I  Q  P  I  C  L  L  N  S  T  Y  K  F  E  N  R  T 140

421 TGACTGCTGGGTGACCGGCTGGGGGGCTATTGGAGAAGATGAGAGTCTGCCATCTCCCAA
     D  C  W  V  T  G  W  G  A  I  G  E  D  E  S  L  P  S  P  N 160
```

FIGURE 18 (AII)

```
481 CACTCTCCAGGAAGTGCAGGTAGCTATTATCAACAACAGCATGTGTAACCATATGTACAA
      T  L  Q  E  V  Q  V  A  I  I  N  N  S  M  C  N  H  M  Y  K 180

541 AAAGCCAGACTTCCGCACGAACATCTGGGGAGACATGGTTTGCGCTGGCACTCCTGAAGG
      K  P  D  F  R  T  N  I  W  G  D  M  V  C  A  G  T  P  E  G 200

601 TGGCAAGGATGCCTGCTTTGGTGACTCGGGAGGACCCTTGGCCTGCGACCAGGATACGGT
      G  K  D  A  C  F  G  D  S  G  G  P  L  A  C  D  Q  D  T  V 220

661 GTGGTATCAGGTTGGAGTTGTGAGCTGGGGAATAGGCTGTGGTCGCCCCAATCGCCCTGG
      W  Y  Q  V  G  V  V  S  W  G  I  G  C  G  R  P  N  R  P  G 240

721 AGTCTATACCAACATCAGTCATCACTACAACTGGATCCAGTCAACCATGATCCGCAATGG
      V  Y  T  N  I  S  H  H  Y  N  W  I  Q  S  T  M  I  R  N  G 260

781 GCTGCTCAGGCCTGACCCAGTCCCCTTGCTACTGTTTCTTACTCTGGCCTGGGCTTCCTC
      L  L  R  P  D  P  V  P  L  L  L  F  L  T  L  A  W  A  S  S 280
841 TTTGCTGAGGCCTGCCTGAGCCCACACGTGTACGTCACACCTGTGAGGTCAGGGTGTGTC
      L  L  R  P  A                                              285

901 TCTTTTGTATCTTGCTTGCTAATAAACCTGTTAATATTTAAAAAAAAAAAAAAAAAAAA
```

FIG 18B

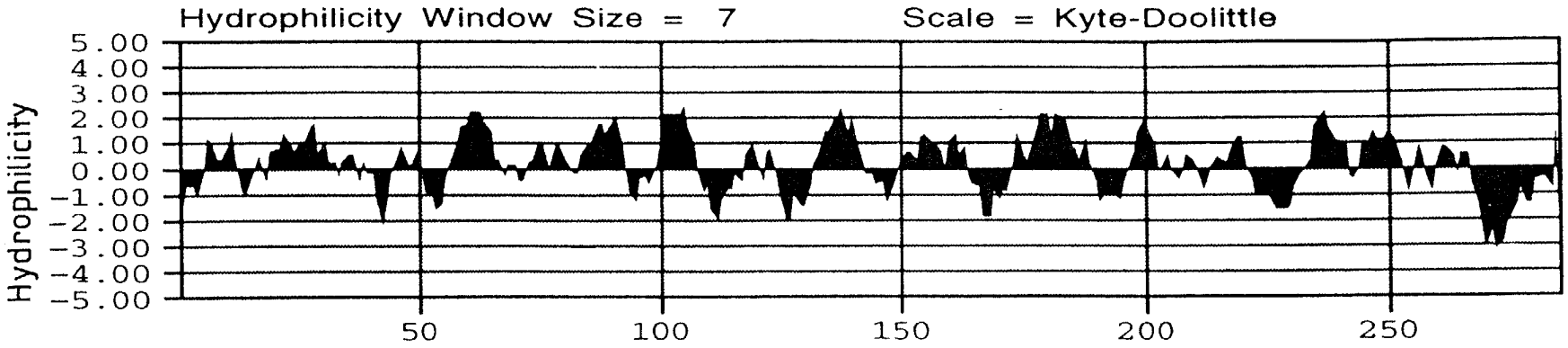
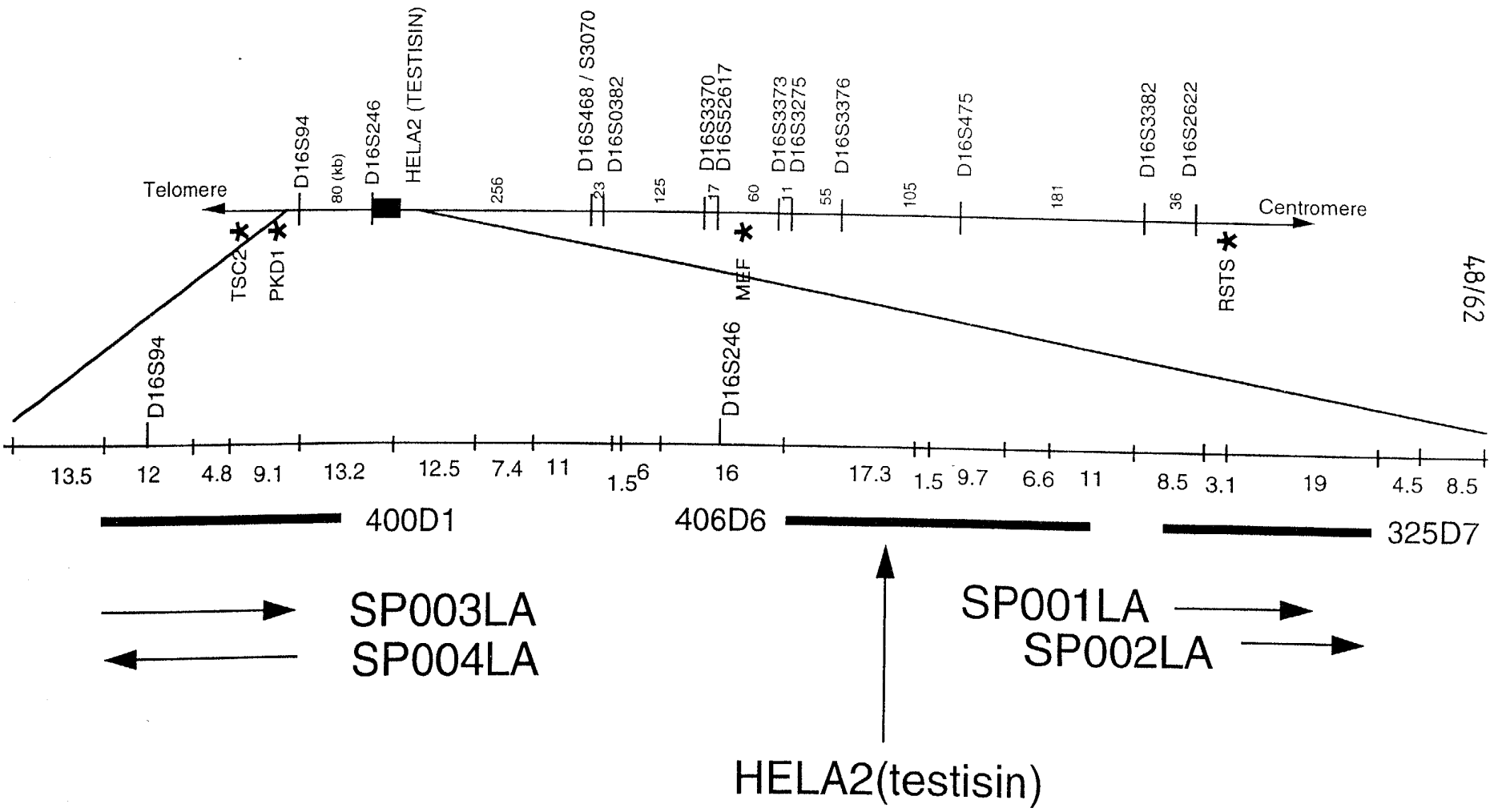


FIGURE 19


Telomere
Centromere
D16S94
D16S246
HELA2 (TESTISIN)
D16S468 / S3070
D16S0382
D16S3370
D16S52617
D16S3373
D16S3275
D16S3376
D16S475
D16S3382
D16S2622
80 (kb)
256
23
125
17
60
1
55
105
181
36
TSC2
PKD1
MEF
RSTS
D16S94
D16S246
13.5
12
4.8
9.1
13.2
12.5
7.4
11
1.5
6
16
17.3
1.5
9.7
6.6
11
8.5
3.1
19
4.5
8.5
400D1
406D6
325D7
SP003LA
SP004LA
SP001LA
SP002LA
HELA2(testisin)

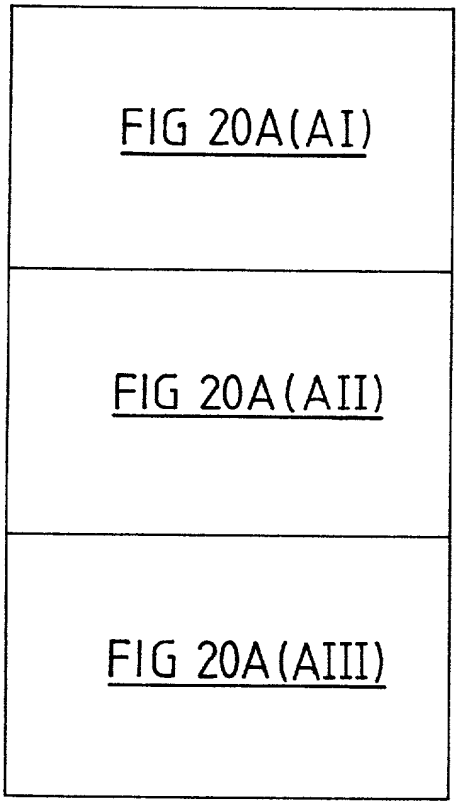


FIG 20A(A)

FIGURE 20A (AI)

```
    CTGAACCGGGTTGTGGGCGGCGAGGACAGCACTGACAGCGAGTGGCCCTGGATCGTGAGC    60
1     L  N  R▽V  V  G  G  E  D  S  T  D  S  E  W  P  W  I  V  S

    ATCCAGAAGAATGGGACCCACCACTGCGCAGGTTCTCTGCTCACCAGCCGCTGGGTGATC   120
21    I  Q  K  N  G  T  H  H  C  A  G  S  L  L  T  S  R  W  V  I

    ACTGCTGCCCACTGTTTCAAGGACAACCTGAACAAACCATACCTGTTCTCTGTGCTGCTG   180
41    T  A  A  Ⓗ  C  F  K  D  N  L  N  K  P  Y  L  F  S  V  L  L

    GGGGCCTGGCAGCTGGGGAACCCTGGCTCTCGGTCCCAGAAGGTGGGTGTTGCCTGGGTG   240
61    G  A  W  Q  L  G  N  P  G  S  R  S  Q  K  V  G  V  A  W  V

    GAGCCCCACCCTGTGTATTCCTGGAAGGAAGGTGCCTGTGCAGACATTGCCCTGGTGCGT   300
81    E  P  H  P  V  Y  S  W  K  E  G  A  C  A  Ⓓ  I  A  L  V  R

    CTCGAGCGCTCCATACAGTTCTCAGAGCGGGTCCTGCCCATCTGCCTACCTGATGCCTCT   360
101   L  E  R  S  I  Q  F  S  E  R  V  L  P  I  C  L  P  D  A  S

    ATCCACCTCCCTCCAAACACCCACTGCTGGATCTCAGGCTGGGGGAGCATCCAAGATGGA   420
121   I  H  L  P  P  N  T  H  C  W  I  S  G  W  G  S  I  Q  D  G
```

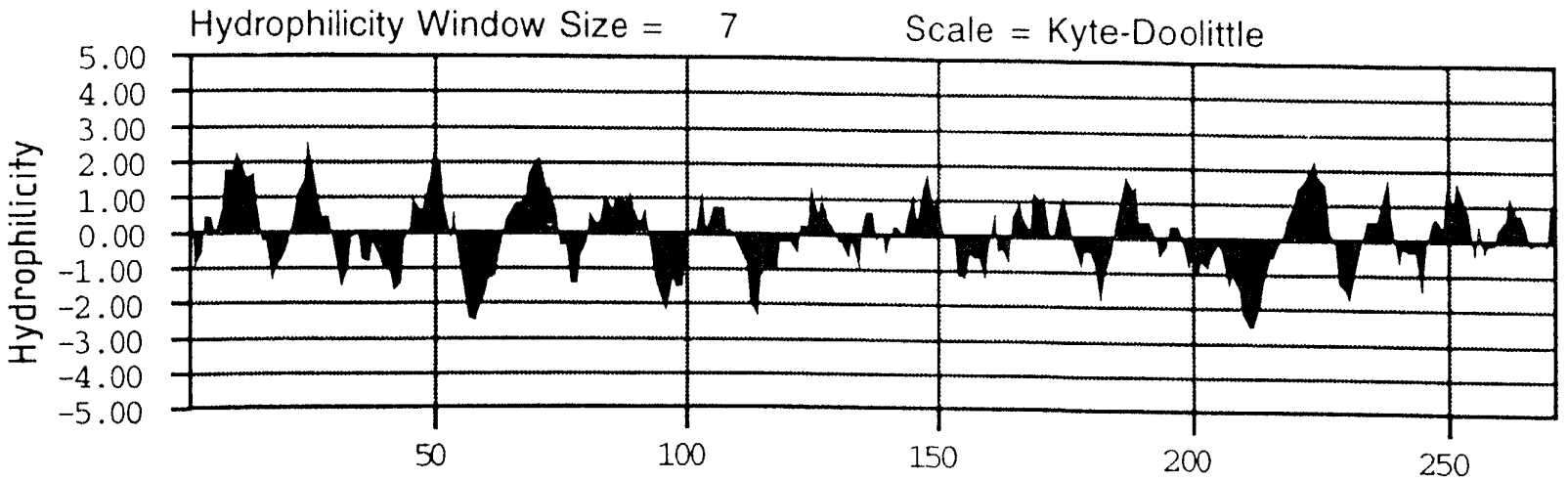
FIGURE 20A (AII)

```
     GTTCCCTTGCCCCACCCTCAGACCCTGCAGAAGCTGAAGGTTCCTATCATCGACTCGGGAA 480
141   V  P  L  P  H  P  Q  T  L  Q  K  L  K  V  P  I  I  D  S  E

     GTCTGCAGCCATCTGTACTGGCGGGGAGCAGGACAGGGACCCATCACTGAGGACATGCTG  540
161   V  C  S  H  L  Y  W  R  G  A  G  Q  G  P  I  T  E  D  M  L

     TGTGCCGGCTAACTTGGAGGGGGAGCGGGATGCTTGTCTGGGCGACTCCGGGGGCCCCCTC 600
181   C  A  G  Y  L  E  G  E  R  D  A  C  L  G  D  S  G  G  P  L

     ATGTGCCAGGTGGACGGCGCCTGGCTGCTGGCCGGCATCATCAGCTGGGGCGAGGGCTGT  660
201   M  C  Q  V  D  G  A  W  L  L  A  G  I  I  S  W  G  E  G  C

     GCCGAGCGCAACAGGCCCGGGGTCTACATCAGCCTCTCTGCGCACCGCTCCTGGGTGGAG  720
221   A  E  R  N  R  P  G  V  Y  I  S  L  S  A  H  R  S  W  V  E

     AAGATCGTGCAAGGGGTGCAGCTCCGCGGGCGCGCTCAGGGGGGTGGGGCCCTCAGGGCA  780
241   K  I  V  Q  G  V  Q  L  R  G  R  A  Q  G  G  G  A  L  R  A

     CCGAGCCAGGGCTCTGGGGCCGCCGCGCGCTCCTAGGGGCCCAGCGGGACGCGGGGCTCGG 840
261   P  S  Q  G  S  G  A  A  A  R  S

     ATCTGAAAGGCGGCCAGATCCACATCTGGATCTGGATCTGCGGCGGCCTCGGGCGGTTTC  900
     CCCCGCCGTAAATAGGCTCATCTACCTCTACCTCTGGGGGCCCGGACGGCTGCTGCGGAA  960
```

FIGURE 20A (AIII)

```
AGGAAACCCCCTCCCCGACCCGCCCGACGGCCTCAGGCCCCGCCTCCAAGGCATCAGGCC 1020
CCGCCCCAACGGCCTCATGTCCCCGCCCCCACGACTTCCGGCCCCGCCCCGGGCCCCAGCG 1080
CTTTTGTGTATATAAATGTTAATGATTTTTATAGGTATTTGTAACCCTGCCACATATCT  1140
TATTTATTCCTCCAATTTCAATAAA
```

FIG 20A (B)


Hydrophilicity Window Size = 7
Scale = Kyte-Doolittle
Hydrophilicity
5.00
4.00
3.00
2.00
1.00
0.00
-1.00
-2.00
-3.00
-4.00
-5.00
50
100
150
200
250

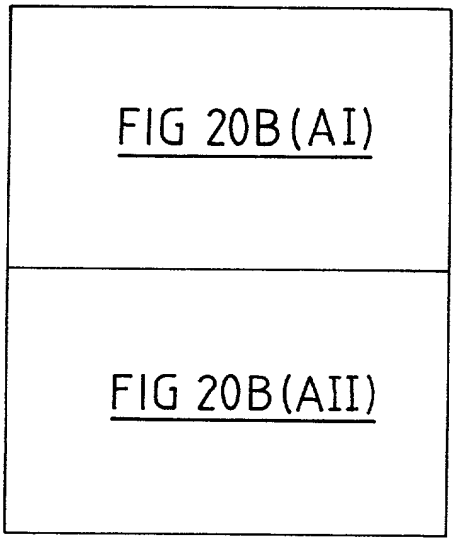


FIG 20B(A)

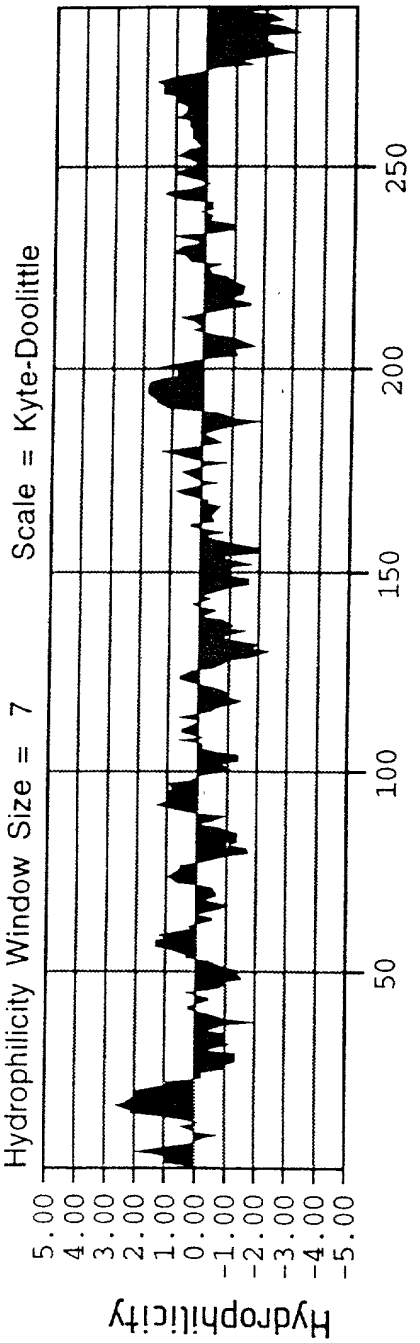
FIGURE 20B (AI)

```
    AATGCGGCCACTCCAAGGAGGCCGGGAGGATTGTGGGAGGCCAAGACACCCAGGAAGGAC  60
1       [C] G  H  S  K  E  A  G  R▽I  V  G  G  Q  D  T  Q  E  G

    GCTGGCCGTGGCAGGTTGGCCTGTGGTTGACCTCAGTGGGGCATGTATGTGGGGGCTCCC 120
21  R  W  P  W  Q  V  G  L  W  L  T  S  V  G  H  V [C] G  G  S

    TCATCCACCCACGCTGGGTGCTCACAGCCGCCCACTGCTTCCTGAGGTCTGAGGATCCCG 180
41  L  I  H  P  R  W  V  L  T  A  A (H)[C] F  L  R  S  E  D  P

    GGCTCTACCATGTTAAAGTCGGAGGGCTGACACCCTCACTTTCAGAGCCCCACTCGGCCT 240
61  G  L  Y  H  V  K  V  G  G  L  T  P  S  L  S  E  P  H  S  A

    TGGTGGCTGTGAGGAGGCTCCTGGTCCACTCCTCATACCATGGGACCACCACCAGCGGGG 300
81  L  V  A  V  R  R  L  L  V  H  S  S  Y  H  G  T  T  T  S  G

    ACATTGCCCTGATGGAGCTGGACTCCCCCTTGCAGGCCTCCCAGTTCAGCCCCATCTGCC 360
101 (D) I  A  L  M  E  L  D  S  P  L  Q  A  S  Q  F  S  P  I [C]

    TCCCAGGACCCCAGACCCCCCTCGCCATTGGGACCGTGTGCTGGGTAAACGGGCTGGGGG 420
121 L  P  G  P  Q  T  P  L  A  I  G  T  V [C] W  V  N  G  L  G

    TCCACTCAGGAGAGGCCCTGGCGAGTGTCCTTCAGGAGGTGGCTGTGCCCCTCCTGGACT 480
141 V  H  S  G  E  A  L  A  S  V  L  Q  E  V  A  V  P  L  L  D
```

FIGURE 20B (AII)

```
    CGAACATGTGTGAGCTGATGTACCACCTAGGAGAGCCCAGCCTGGCTGGCCAGCGCCTCA 540
161 S   N   M   C   E   L   M   Y   H   L   G   E   P   S   L   A   G   Q   R   L

    TCCAGGACGACATGCTCTGTGCTGGCTCTGTCCAGGGCAAGAAAGACTCCTGCCAGGGTG 600
181 I   Q   D   D   M   L   C   A   G   S   V   Q   G   K   K   D   S   C   Q   G

    ACTCCGGGGGGCCGCTGGTCTGCCCCATCAATGATACGTGGATCCAGGCCGGCATTGTGA 660
201 D   S   G   G   P   L   V   C   P   I   N   D   T   W   I   Q   A   G   I   V

    GCTGGGGATTCGGCTGTGCCCGGCCTTTCCGGCCTGGTGTCTACACCCAGGTGCTAAGCT 720
221 S   W   G   F   G   C   A   R   P   F   R   P   G   V   Y   T   Q   V   L   S

    ACACAGACTGGATTCAGAGAACCCTGGCTGAATCTCACTCAGGCATGTCTGGGGCCCGCC 780
241 Y   T   D   W   I   Q   R   T   L   A   E   S   H   S   G   M   S   G   A   R

    CAGGTGCCCCAGGATCCCACTCAGGCACCTCCAGATCCCACCCAGTGCTGCTGCTTGAGC 840
261 P   G   A   P   G   S   H   S   G   T   S   R   S   H   P   V   L   L   L   E

    TGTTGACCGTATGCTTGCTTGGGTCCCTGTGAACCATGAGCCATGGAGTCCGGGATCCCC 900
281 L   L   T   V   C   L   L   G   S   L

    TTTCTGGTAGGATTGATGGAATCTAATAATAAA
```

FIG 20B (B)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Scale = Kyte-Doolittle
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4.00
3.00
2.00
1.00
0.00
-1.00
-2.00
-3.00
-4.00
-5.00
50
100
150
200
250

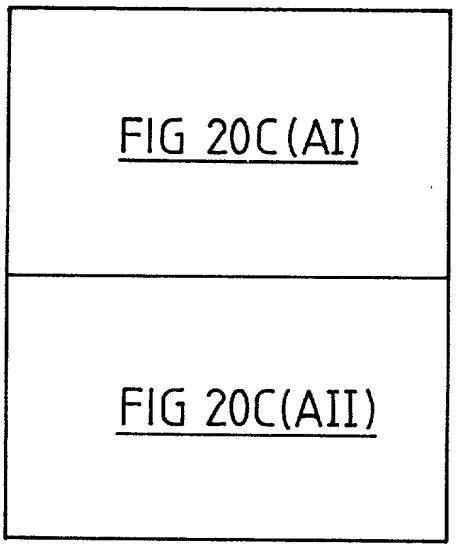


FIG 20C(A)

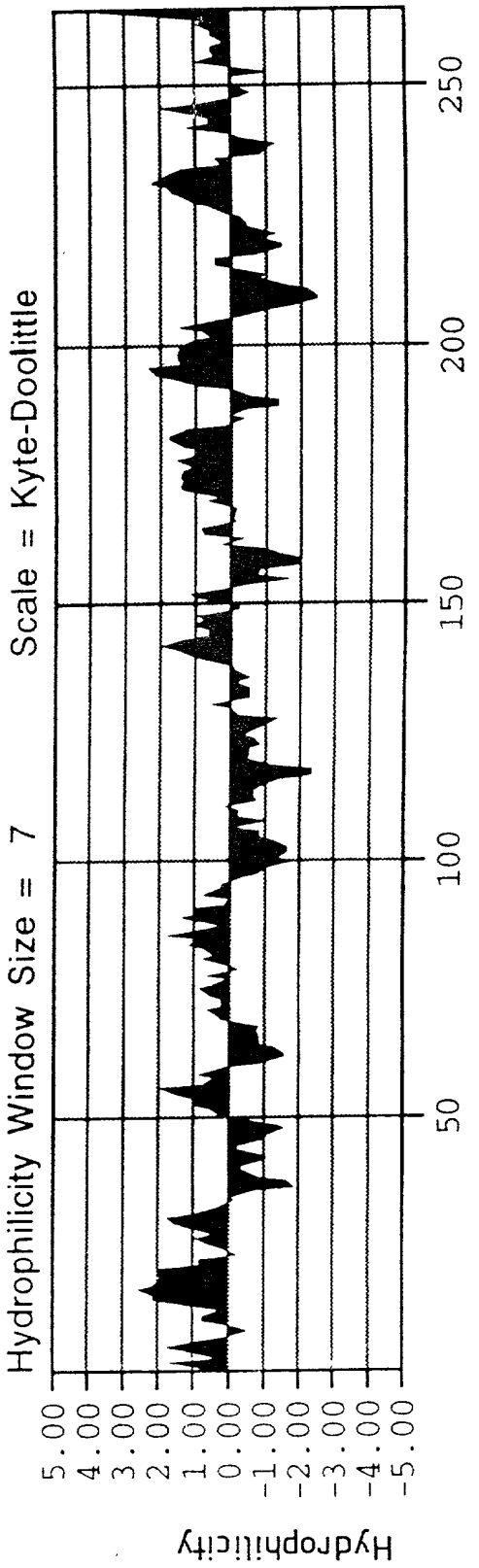
FIGURE 20C (AI)

```
    CCTGTGGTCGCCCCAGGATGCTGAACCGAATGGTGGGCGGGCAGGACACGCAGGAGGGCG  60
1     [C] G  R  P  R  M  L  N  R▽M  V  G  G  Q  D  T  Q  E  G

    AGTGGCCCTGGCAAGTCAGCATCCAGCGCAACGGAAGCCACTTCTGCGGGGGCAGCCTCA 120
21  E  W  P  W  Q  V  S  I  Q  R  N  G  S  H  F [C] G  G  S  L

    TCGCGGAGCAGTGGGTCCTGACGGCTGCGCACTGCTTCCGCAACACCTCTGAGACGTCCC 180
41  I  A  E  Q  W  V  L  T  A  A (H)[C] F  R  N  T  S  E  T  S

    TGTACCAGGTCCTGCTGGGGGCAAGGCAGCTAGTGCAGCCGGGACCACACGCTATGTATG 240
61  L  Y  Q  V  L  L  G  A  R  Q  L  V  Q  P  G  P  H  A  M  Y

    CCCGGGTGAGGCAGGTGGAGAGCAACCCCCTGTACCAGGGCACGGCCTCCAGCGCTGACG 300
81  A  R  V  R  Q  V  E  S  N  P  L  Y  Q  G  T  A  S  S  A (D)

    TGGCCCTGGTGGAGCTGGAGGCACCAGTGCCCTTCACCAATTACATCCTCCCCGTGTGCC 360
101 V  A  L  V  E  L  E  A  P  V  P  F  T  N  Y  I  L  P  V [C]

    TGCCTGACCCCTCGGTGATCTTTGAGACGGGCATGAACTGCTGGGTCACTGGCTGGGGCA 420
121 L  P  D  P  S  V  I  F  E  T  G  M  N [C] W  V  T  G  W  G

    GCCCCAGTGAGGAAGACCTCCTGCCCGAACCGCGGATCCTGCAGAAACTCGCTGTGCCCA 480
141 S  P  S  E  E  D  L  L  P  E  P  R  I  L  Q  K  L  A  V  P
```

FIGURE 20C (AII)

```
    TCATCGACACACCCAAGTGCAACCTGCTCTACAGCAAAGACACCGAGTTTGGCTACCAAC 540
161 I   I   D   T   P   K   C   N   L   L   Y   S   K   D   T   E   F   G   Y   Q

    CCAAAACCATCAAGAATGACATGCTGTGCGCCGGCTTCGAGGAGGGCAAGAAGGATGCCT 600
181 P   K   T   I   K   N   D   M   L   C   A   G   F   E   E   G   K   K   D   A

    GCAAGGGCGACTCGGGCGGCCCCCTGGTGTGCCTCGTGGGTCAGTCGTGGCTGCAGGCGG 660
201 C   K   G   D   S   G   G   P   L   V   C   L   V   G   Q   S   W   L   Q   A

    GGGTGATCAGCTGGGGTGAGGGCTGTGCCCGCCAGAACCGCCCAGGTGTCTACATCCGTG 720
221 G   V   I   S   W   G   E   G   C   A   R   Q   N   R   P   G   V   Y   I   R

    TCACCGCCCACCACAACTGGATCCATCGGATCATCCCCAAACTGCAGTTCCAGCCAGCGA 780
241 V   T   A   H   H   N   W   I   H   R   I   I   P   K   L   Q   F   Q   P   A

    GGTTGGGCGGCCAGAAGTGAGACCCCCGGGGCCAGGAGCCCCTTGAGCAGAGCTCTGCAC 840
261 R   L   G   G   Q   K   *   D   P   R   G   Q   E   P   L   E   Q   S   S   A

    CCAGCCTGCCCGCCCACACCATCCTGCTGGTCCTCCCAGCGCTGCTGTTGCACCTGTGAG 900
281 P   S   L   P   A   H   T   I   L   L   V   L   P   A   L   L   L   H   L

    CCCCACCAGACTCATTTGTAAATAGCGCTCCTTCCTCCCCTCTCAAATACCCTTATTTTA 960
    TTTATGTTTCTCCCAATAAA
```

FIG 20C(B)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Scale = Kyte-Doolittle
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4.00
3.00
2.00
1.00
0.00
-1.00
-2.00
-3.00
-4.00
-5.00
50
100
150
200
250

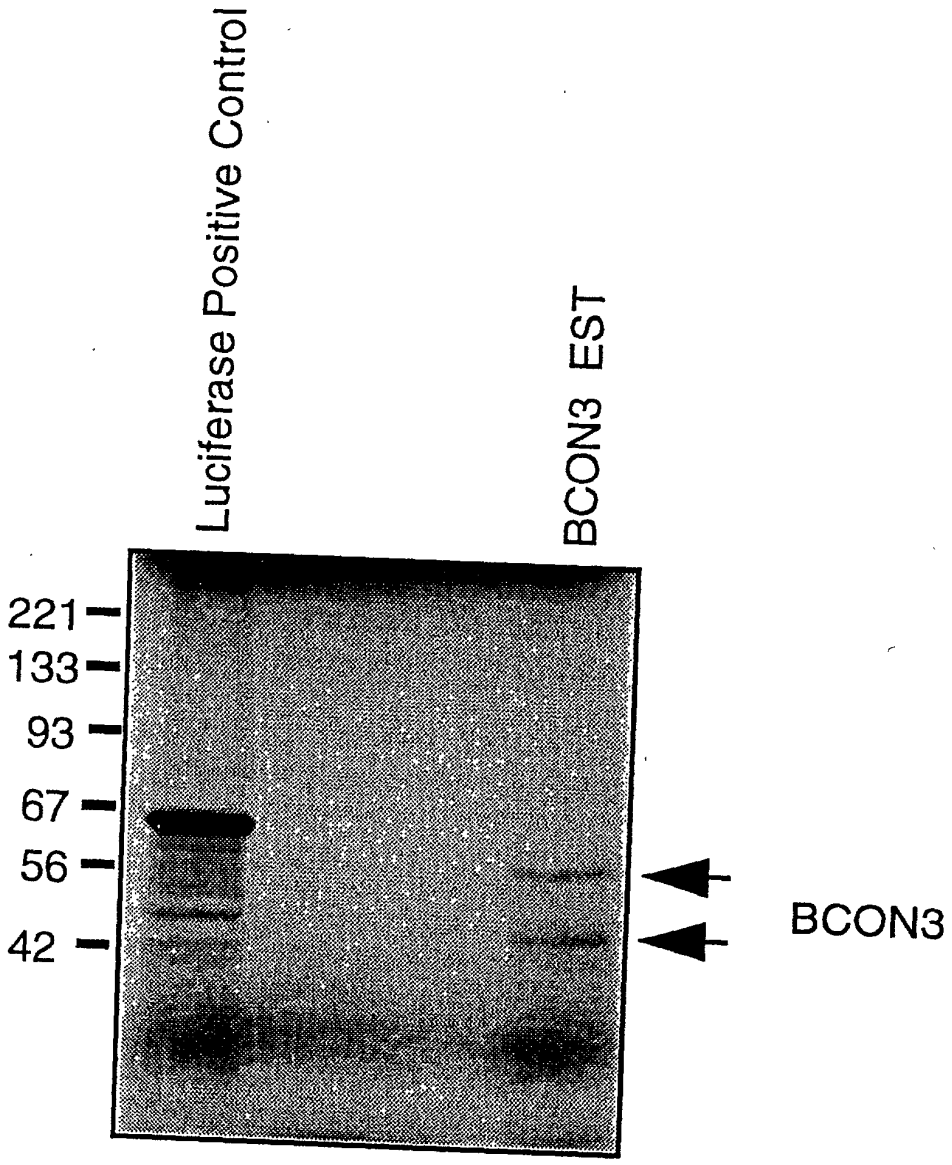
Luciferase Positive Control
BCON3 EST
221
133
93
67
56
42
BCON3

FIG 21